NASA SP-64

PROGRESS IN DEVELOPMENT OF METHODS IN BONE DENSITOMETRY

A conference held in Washington, D. C., March 25-27, 1965, and sponsored by National Aeronautics and Space Administration, National Institute of Arthritis and Metabolic Diseases (NIH), and American Institute of Biological Sciences

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Foreword

The effects of weightlessness and decreased activity on the astronaut's musculoskeletal system during prolonged space flight missions are of concern to NASA. This problem was anticipated from the knowledge that human subjects lose significant quantities of calcium from the skeleton during periods of bedrest, immobilization, and water immersion.

An accurate method of measurement of the changes in mineral content of the skeleton is required not only in the space program but also in the biological, medical, and dental fields for mineral metabolism studies and for studying various pathological conditions of the skeleton and teeth. This method is a difficult one requiring the coordinated efforts of physiologists, biophysicists, radiologists, and clinicians. The densitometry methods reported in this conference which have been used or are being developed include X-ray, beta excited X-rays, radioisotopes, sonic vibration, and neutron activation analysis. Studies in the Gemini, Biosatellite, and Apollo flights use the X-ray bone densitometry method which requires making X-rays before and after the flights. An in-flight method of bone densitometry would be valuable, and use of radioisotope sources has been suggested.

Many advances in bone densitometry have been made in the last five years, and the urgency of the requirement makes this working conference timely and valuable. In such a rapidly developing field with investigators working independently in a variety of scientific disciplines, a working conference is of great value in exchanging information and ideas, critically evaluating approaches and methods, and pointing out new research pathways.

This working conference was organized under the joint sponsorship of the National Institute of Arthritis and Metabolic Diseases of the National Institutes of Health, the American Institute of Biological Sciences, and the National Aeronautics and Space Administration. These proceedings are the written record of the invited papers and informal discussion. The editors have deleted much of the discussion involving clinical problems, bone pathology, and other areas not directly related to bone densitometry. Lengthy discussions on X-ray densitometry were deleted since the major controversies on linearization, analog transformation, and instrumentation problems were resolved. This working conference was of great value in making a critical evaluation of the present status of methods in bone densitometry and in stimulating further interest in this research area.

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Chief, Environmental Biology
Office of Space Science and Applications
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Welcoming Remarks

DALE W. JENKINS, Conference Chairman
JOHN R. OLIVE, American Institute of Biological Sciences

DR. JENKINS:

On behalf of the National Aeronautics and Space Administration, I would like to welcome the participants to this Working Conference on Progress in Development of Methods in Bone Densitometry.

Dr. G. Donald Whedon and I have discussed the importance and the need of getting research people together in this field to determine what progress has been made. There have been many developments in bone densitometry using X-ray, radioisotopes, and sonic vibration. We have asked the principal scientists in the world working in this area to come together with a critical group of knowledgeable specialists in related fields.

The field of calcium metabolism is so controversial and difficult that a working conference of this type is quite necessary. When a group of calcium specialists and densitometry people get together, you get some controversy, and occasionally a little excitement. Now that we have a critical group, maybe we will have a critical mass, perhaps leading to an explosive advance.

The problem of bone densitometry in humans and animals has existed for a long time, and it has medical, dental, and public health importance. With the advent of the space age, it assumes greater importance with regard to astronauts under weightlessness conditions. Based on results from bed rest, immobilization, and water immersion studies, it has been anticipated that decalcification in astronauts during long periods of weightlessness will be a potential problem.

Bone X-ray densitometry experiments are scheduled on several of the Gemini flights to determine any change in bone mineral content of the astronauts. It is essential that the present status of the improved X-ray measurement method be fully understood and points of controversy be impartially discussed. It is hoped that this working conference can resolve some of the questions and problems which have been associated with the earlier attempts to use bone X-ray densitometry. The proceedings of this conference will provide a timely reference and critical evaluation of this method at the time when the Gemini experiment data are available.

The conference will be very informal, with perhaps one rule that Dr. Frank Fremont-Smith makes in his conferences, "Don't interrupt me while I am interrupting you."

This conference is being co-sponsored by NASA, by the National Institute of Arthritis and Metabolic Diseases (of NIH), and by the American Institute of Biological Sciences (AIBS).

DR. OLIVE:

I would like to add the welcome of the American Institute of Biological Sciences to those of the other co-sponsors.

This is just a brief word about where AIBS fits into this particular program, and its actual inception. We work closely with NASA on several projects, this being a typical example of

how we are able to help coordinate and assemble the scientists.

Realizing that the interests of AIBS are quite broad in the biological medical sciences, it is not difficult, then, to see why we would be interested in co-sponsoring and working with a program of this type. Our own charter is broad, very broad actually, encompassing all the biological sciences as well as the medical and agricultural sciences.

We are an organization now of about 13,000 individuals, with an affiliate membership of about 65,000, and accomplish many of the things the Federation (Federation of American Societies for Experimental Biology) does. We have the same sort of active programs, our own annual meetings, and are interested in the educational side of the biological sciences.

Again, back to NASA for a moment. We have several contracts with NASA on which we are working actively for the furtherance of space sciences as we refer to it, because it includes the manned orbital laboratories and the Biosciences Program of NASA.

Introduction

G. Donald Whedon

First of all, I want to indicate what should be obvious, the strong interest of the National Institutes of Health and particularly of my own Institute, Arthritis and Metabolic Diseases, in this subject. Increasing interest in the problem of bone densitometry has been apparent to us at NIH as it is to everyone else, so that I enjoyed participating with Dale Jenkins in the instigation of this conference.

I think that the Gemini and Mercury launchings certainly point up the importance of developing the most efficient and accurate tool for measuring changes in the density of the skeleton. Nevertheless, for all of our concern about what is going to happen to the skeleton under the condition of weightlessness, bone densitometry has been a problem of great interest over a long period of time to physiologists and clinicians with regard to the effects of various factors on the skeleton, be they hormonal, nutritional, or whatever.

Of course, one of the principal methods that we have used for measuring skeletal changes over some considerable period of time has been the metabolic balance approach. But, even metabolic balance studies for calcium over several weeks' time, or even several months' time, give only a portion of the total biological situation. We need, obviously, some index, some means of measuring very precisely the state of mineralization of various parts of the skeleton from one point in time to another, not only over many months, but over many years, and with real accuracies.

The problems of bone densitometry, I think, have been long known to many scientists. The first papers on this subject appeared at least 30 to 40 years ago. Since that time, there have been comments on the effects of the varying kilovoltage exposure of the films, quality and kind of films, and the very important problem of soft tissue interference, which itself varies a great deal in its density from point to point across any particular bone. The matter of integration by scanning, then, has been of concern, and is a relatively recent development in this field.

An idea of the difficulty of the densitometry problem can be gained from the fact that there has been a tendency to measure those bones which are easiest to measure, with lack of full awareness of the different physiological significance of different bones. In other words, it is much easier to measure the thickness of cortical bone, particularly if it is a very peripheral bone, and a small bone, because it is covered with the least amount of soft tissue. On the other hand, as we are progressively becoming more and more aware, these peripheral bones may be far less responsive to changes in the total mineral content of the skeleton than trabecular bone, which is usually located in spots which are least accessible to accurate X-ray viewing.

The mobility or availability of the mineral in trabecular bone, in apparent contrast to that in cortical bone, may be indicated by two examples. One is the graphic comparison by Vose (Vose et al., 1961. Quantitative Bone Strength Measurements in Senile Osteoporosis. J. Gerontol., vol. 16, p. 120) of a normal femur in cross section, showing the rather dense and convoluted trabecular structure within the cortex, with a similar cross section of a femur from an individual of similar age with pronounced osteoporosis. There is a vast difference in trabecular content of these two bones, with much less difference, at least to the naked eye, with respect to thickness of cortex.

My other example is merely to remind you of the experiment of Bauer (Bauer et al., 1929. A Study of the Bone Trabeculae as a Readily Available Reserve Supply of Calcium. J. Exptl. Med., vol. 49, p. 145). This demonstrated rather graphically the reservoir capacity of trabecular bone for making available the shift of rather large amounts of calcium in and out of the skeleton. These investigators gave one group of adult cats a rather large calcium intake over a period of weeks and months, then they removed one humerus. This same group was then shifted to a low calcium intake for a similar period of time, and finally the other humerus was removed for comparison. Abundant trabecular bone was noted in the humerus from the high calcium feeding period, and later in the same cat, loss of considerable trabecular structure on low calcium. To control the experiment, the investigators took another group of adult cats in which the reverse order of feeding was used, with low calcium intake first, which resulted in fewer and thinner trabeculae. After several weeks of high calcium intake, replacement of trabecular structure in a significant degree was noted. Similar studies were also continued by this group in other animals, and at different ages, but I think the significant point for us is the viability of trabecular bone in adult animals.

In some respects, this is the second workshop conference on methods of bone densitometry. Stanley Garn organized a workshop which was held at NIH in 1959. This one-day conference resulted in a rather excellent bibliography which has been very useful. Due to the considerable interest and activity over the last few years, we felt that it was time to call together a conference which would try to come to grips with this subject with greater vigor, and spend more time at it. This will be a working conference, in which we expect to learn from each other.

We have purposely selected a group from many different disciplines, radiologists, pathologists, clinicians, biophysicists; and there are several others, including mathematicians, who are prepared to make comments on some of our methodology from their point of view.

X-RAY DENSITOMETRY

G. DONALD WHEDON, *Chairman*

Theoretical Aspects of Radiographic Densitometry

S. David Rockoff, Department of Radiology, National Institutes of Health

Quantitative radiographic densitometry has been one of the most thoroughly investigated nondestructive methods for the determination of bone mineral *in vivo*. In evaluating this method, it is well for us to consider some of the sources of systematic bias which may occur and are often overlooked.

The top of figure 1 shows a frequency distribution curve of a typical heterogeneous X-ray beam. The range of X-ray energies is very wide, only a small portion being of the peak energy.

If this X-ray beam strikes a piece of bone, represented by this figure, the heterogeneous radiation is absorbed in a complex fashion. That is, as the heterogeneous beam passes through the bone, not only is the total amount of radiation changed but so is the frequency distribution of the remaining photons, the lower energy photons being absorbed to a greater extent than the higher energy ones. It would be convenient if X-ray beams were homogeneous, since their absorption would then follow a more predictable pattern, and quantitation would be greatly simplified. But the production of homogeneous X-ray beams is difficult and has not yet been developed to a practical, useful level.

It is the usual practice, when X-raying parts of the body for radiographic quantitation, to expose a wedge of known composition to the X-ray beam simultaneously with the body part being studied, in order to have a frame of reference for calibration. Thus, the mean optical

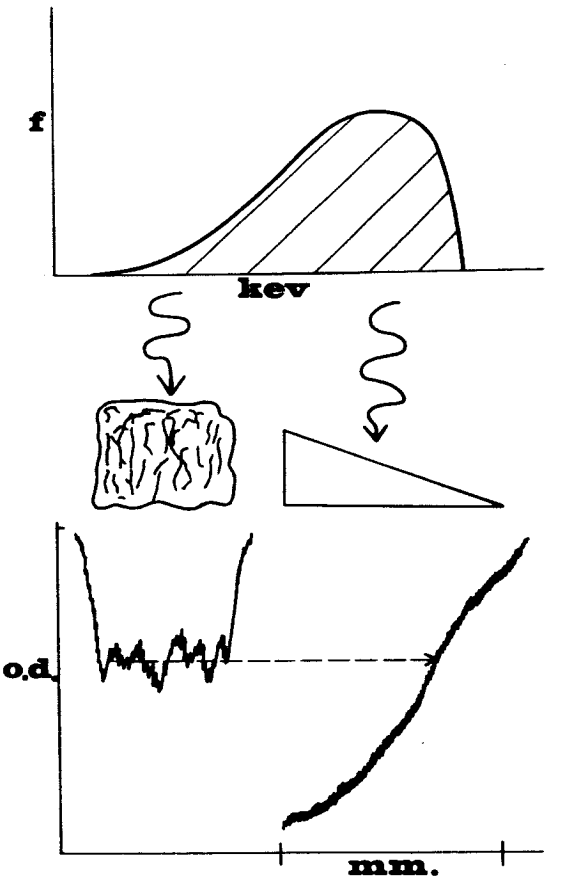

FIGURE 1.—*Top:* Frequency distribution of a typical heterogeneous X-ray beam. *Middle:* Diagrammatic representation of bone (left); calibration wedge (right). *Bottom:* Densitometric scan lines across bone (left) and the calibration wedge (right), showing the manner in which the mean optical density of bone is expressed in terms of wedge thickness.

density of the bone, obtained from densitometric scanning of the radiographic image of the bone, is then expressed in terms of thickness of calibration wedge material which gives the same optical density.

So far, the experimental procedure appears to be straightforward. The end result of the procedure depends upon the film response curve. The factors affecting this curve are probably among the least understood aspects of radiographic densitometry, and may well be the source of discrepancies among investigators. Data are presented which may help clarify some of the properties of the film response curve to help answer the question, "What factors in radiographic densitometry does the film response curve allow us to vary and still obtain unbiased results?"

First, does using a calibration wedge when X-raying a bone allow us to change the field size and still obtain the same results?

Figure 2 shows data obtained by exposing a 15-cm thick presswood phantom and an aluminum wedge simultaneously to X-rays, using X-ray beams of 30, 60, and 90 KVP, and varying the field size between 13 and 60 cm in diameter. The marked difference in the millimeter of aluminum equivalency of the phantom obtained at any given kilovoltage, due only to the changes in field size, is readily seen. In addition, there is a marked difference in results obtained by using X-ray beams of different peak energies. Changes in field size and in kilovoltage result in alterations in the apparent amount of mineral present.

It is generally accepted that different results are also obtained if different types of X-ray films are used, if different types of film holders are used (for instance, if intensifying screens are used instead of cardboard holders), or if there are marked variations in the processing technique. Therefore, for a calibration wedge to be an effective and reliable standard, it appears that the field size, kilovoltage, film type, film holder, and film processing technique should be kept constant.

Figure 3 shows for what factors the film response curve compensates. If a 15-cm phantom and calibration wedge are exposed simultaneously, and the overall densities of the films are arbitrarily changed simply by changing the duration of the exposures, it is noted that for any given X-ray beam between 30 and 90 KVP the wedge equivalency of the phantom is essentially unchanged, no matter how light or dark the films may be. Theoretically, therefore, there is no reason to make films very light or very dark, from the quantitation standpoint, since the same results are obtained in each instance.

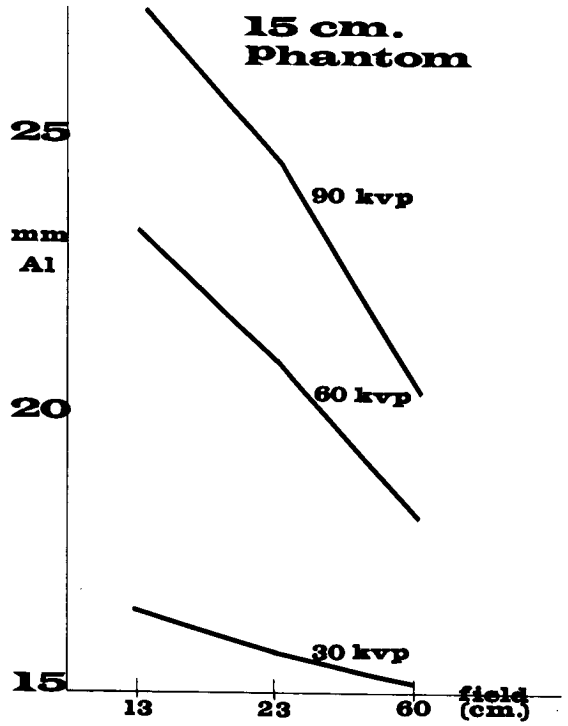

FIGURE 2.—Millimeter of aluminum equivalent of a 15-cm thick phantom, using X-ray field sizes ranging from 13 to 60 cm in diameter, and X-ray energies of 30, 60, and 90 KVP. Note that the size of the field and the X-ray energy level markedly affect the millimeter of aluminum equivalence of the phantom.

Is there, in fact, a linear relationship between the thickness of (or amount of mineral in) a piece of bone and the equivalent amount of wedge material in which it is expressed? Let us consider why it is desirable to have a linear relationship present. Consider two situations, one in which there is a linear relationship between the amount of mineral and wedge equivalency, and one in which there is not. In the first

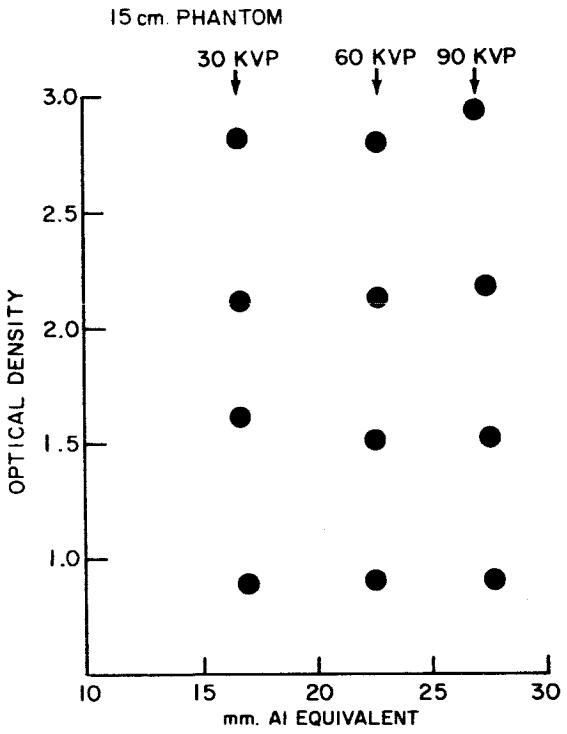

FIGURE 3.—Plot of millimeter of aluminum equivalence of a 15-cm phantom, and the darkness of the films analyzed, as expressed in optical density. Tests were done at 30, 60, and 90 KVP. Note that the darkness of the film, at any particular X-ray energy, has no significant effect on the millimeter of aluminum equivalence.

instance, there is a uniformly proportional relationship present. That is, a given increment in wedge material corresponds to a constant increment of bone mineral at every level. In the case of the nonlinear relationship, however, the same increments in wedge equivalency correspond to different increments of bone mineral, depending upon where on the curve the data points occur.

When phantom thicknesses are plotted against millimeters of aluminum equivalency (fig. 4), there is a generally linear relationship at 30, 60, and 90 KVP when a small X-ray field (13 cm in diameter) is used. When a large X-ray field is used, however, this linear relationship (indicated by the broken lines) is apparently lost. The fact that the elbows of the curves are always in the downward direction suggests that these are real aberrations and are not due to random variation. This loss of linearity with *large* X-ray fields demonstrates the importance of using *small* X-ray fields when doing quantitative densitometry.

These are some of the sources of systematic bias which may affect the film response curve and the relationship of this curve to apparent mineral content. I would like to turn to what may be done to the film response curve itself to increase precision and convenience, while still

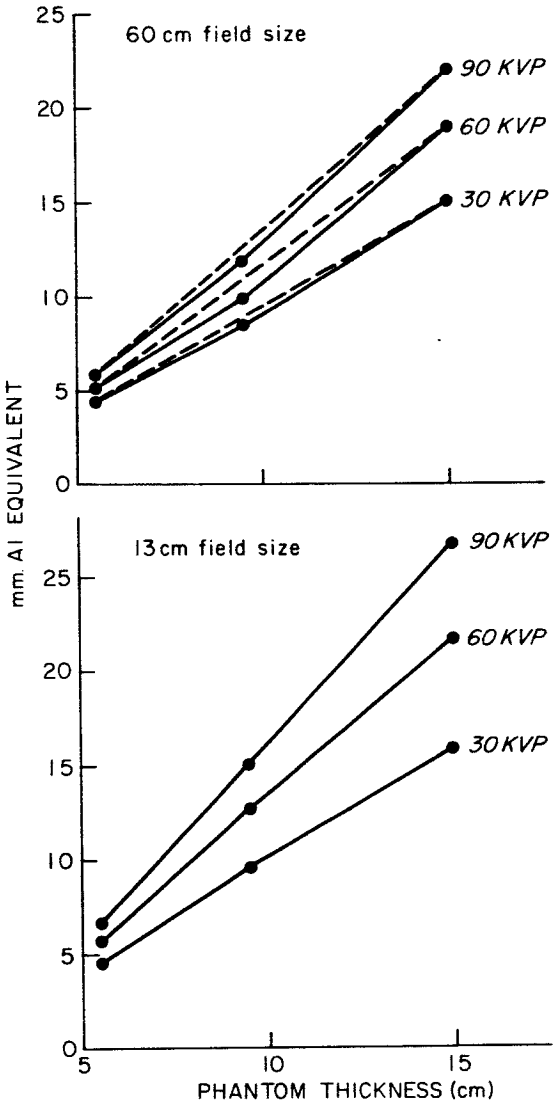

FIGURE 4.—Plot of millimeter of aluminum equivalents and phantom thickness for a 13-cm diameter field (bottom) and a 60-cm diameter X-ray field (top). Tests were done at 30, 60, and 90 KVP. Note that with the large X-ray field, the linear relationship present with the smaller X-ray field is lost.

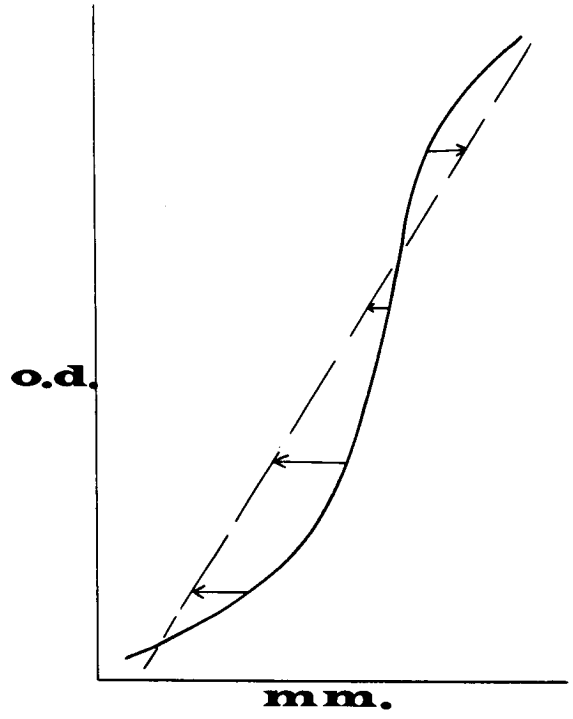

FIGURE 5.—Typical film response curve (optical density vs. millimeter thickness of calibration wedge) is represented by the solid line. The bias which is introduced by arbitrarily altering the shape of this curve is demonstrated by the broken line and the arrows. This bias does not apply to a transformation of the curve, the transforms of which are also applied to the bone scan line.

keeping it a reliable index of the amount of mineral present in the specimen being studied.

In Figure 5 the solid line represents a film response curve obtained by scanning densitometry of the radiographic image of the calibration wedge (the plot of the optical density vs. the millimeter thickness of the wedge). It is quite permissible to perform the procedures upon the curve which would be necessary to smooth the curve locally, that is, we may draw a smooth curve through the random fluctuations which occur due to film grain and electronic noise. Linearization of the curve over its entire length, with subsequent expression of bone density in terms of the new line, is not permissible since this erroneously gives the same precision over the entire range of optical densities and introduces a varying bias into the results. On the other hand, expressing the area under a bone scan curve in terms of a transformed film response curve is permissible, *if* the same transforming procedures are performed point-to-point on the bone scan values as were performed on the film response curve. In figure 6 is shown the scanning densitometric and recording instrumentation in use in this laboratory.

These introductory remarks about the nature of the film response curve and some of the factors which affect it should serve as useful background material.

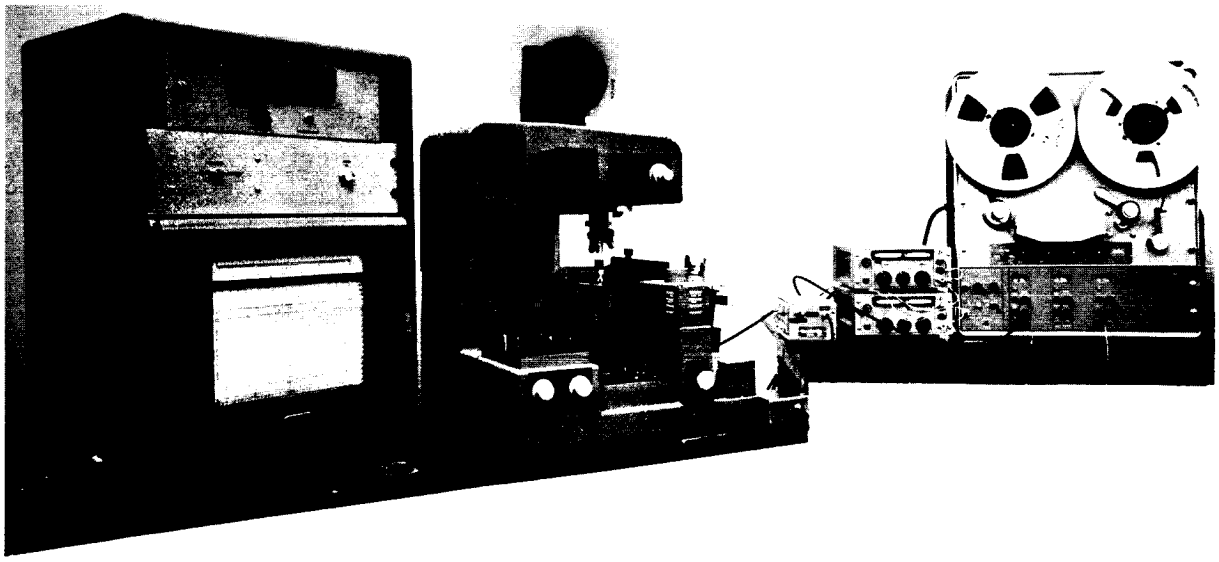

FIGURE 6.—New, versatile scanning and recording instrumentation in use in this laboratory.

Quantitative Radiography of the Skeleton in Living Systems

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The discovery of X-rays by Roentgen in 1895 probably ranks as one of the great scientific discoveries. Within three months of the discovery, X-rays were being put to practical use in a Viennese hospital in connection with surgery. The history of X-ray physics illustrates the importance to the entire world of research in pure science. Had Roentgen deliberately set about to discover some means of assisting surgeons in reducing fractures, it is almost certain that he would never have been working with the evacuated tubes, induction coils, and the like, which led to his famous discovery (Richtmeyer et al., 1955).

One of the many possible biomedical applications of this discovery is the accurate assessment of the bone mineral content of a living subject. The need for such a method is clearly evident by the many investigations in this area. The successful development of such a method would assist significantly in the detection of normal skeletal variation, in the recognition of the incipient or latent stages of pathological conditions involving bone, or in the early responses of osseous disorders to therapy.

A practical quantitative technique is described for the measurement and evaluation of bone mass and bone density from roentgenograms. Evidence is presented that the use of this procedure provides data from animals and man which correlate with physical and chemical properties of bone, and with alterations of physiological and pathological significance in the skeletal system.

The method in use at Pennsylvania State University has gone through a long period of development beginning in 1927 under the aegis of P. B. Mack. An apparatus was developed by Brown (1949) which greatly facilitated the extraction of information from suitably exposed and standardized roentgenograms. This apparatus was referred to as a bone density computing machine and formed the basis of the present equipment which was developed in 1957 during my supervision of the laboratory.

THE X-RAY PROCEDURE

Any medical X-ray machine which is operable in the 50 to 60 kv range may be used. A small focal spot is desirable (we use a 1 mm focal spot). No-screen X-ray film in cardboard holders is employed. Intensifier screens are avoided because the scattering effect of the crystals diffuses the image, thereby reducing the sharpness of detail. In addition, different screens may vary in the size of the crystals, and image clarity may differ from laboratory to laboratory (Files, 1959).

A specially designed, standardized aluminum-zinc alloy wedge (fig. 1) is simultaneously exposed with the anatomical part of interest. This wedge is of uniform composition, is mechanically stable, easily machined, and is a good analogue of bone. It contains 92.8% aluminum and 7.2% zinc. A mass of this alloy was produced by the Aluminum Company of America in one melt to provide a large quantity of homogeneous material. A sufficient supply of

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this material is on hand so that many wedges may be fabricated in the interest of universal standardization. Several wedges are presently on loan to investigators in different parts of the world.

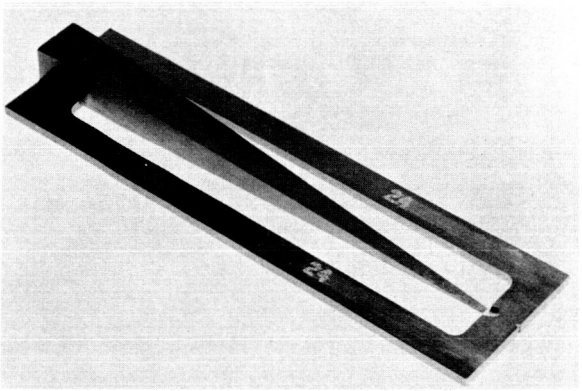

FIGURE 1.—The aluminum alloy wedge which is used as the standard is 14 cm long, 14 mm in width and depth at the thick end, and is 2 mm wide at the tip. The slope is 0.1.

Roentgenograms of the bone to be evaluated are taken according to a technique which is standardized for all possible factors, such as positioning of subject and the wedge, exposure factors, film processing, and other factors. Typical exposure factors for an adult hand would be 55 kv peak and 18 milliampere seconds (MAS) using a 2 mm aluminum filter. The MAS is varied with tube to film distance and mass of the bone. The carefully developed film is then analyzed in the densitometric equipment.

BONE DENSITY MEASURING EQUIPMENT

The densitometer consists of a Leeds and Northrup Microphotometer, a Speedomax Recorder (Type G—Model S6000), and an electronic analogue computer (fig. 2). The tungsten scanning lamp of the microphotometer projects a beam of white light through the radiograph. The intensity of the transmitted light is proportional to the optical density of the radiograph which is, in turn, proportional to the X-ray absorbing properties of the tissue. This light intensity is transformed by a photomultiplier into an electromotive force (emf) which drives the recorder and its balancing circuit. The position of the slide wire contact in this circuit indicates the magnitude of the emf and controls the position of the pen, which records on a chart. The relationship between the chart speed and the film scanning speed is known and controlled.

The X-ray image of the wedge is scanned within prechosen film density limits and its X-ray absorption curve is plotted by the recorder. Ten coordinates of the curve are then set on ten computer potentiometers which impress the X-ray absorption curve of the wedge as a potential curve on a ten-tap helipot mechanically attached to the recorder shaft. In this way the voltage drop of the helipot at any instant is a function of both the pen position and the X-ray absorption curve of the wedge. The bone and soft tissue image is then placed in position and scanned along a selected line, called a trace path. An instantaneous comparison is thus made of the X-ray absorption of a small section of tissue, determined by the area of the scanning beam, with the equivalent X-ray absorption thickness of the wedge. The equivalent wedge thickness potentials previously imposed on the helipot are integrated over the time it takes to scan the tissue. This integrated potential is stored in a capacitor and is proportional to the total mass of absorbing material scanned. When the tracing of the bone and soft tissue image has been completed, the stored potential is balanced against a 10-turn potentiometer with a 3-digit dial. The resulting count is directly proportional to the mass of the tissue whose radiograph was scanned. This value is termed the X-ray mass coefficient after it has been adjusted for equipment calibration, scanning speed, and other mechanical variables. By closely approximating the cross-sectional area of the scanned section, the X-ray density coefficient can be computed.

COMPUTATION OF X-RAY MASS COEFFICIENT

The rat femur will be used as a model for discussing the method of bone mineral content analysis and some of the associated problems.

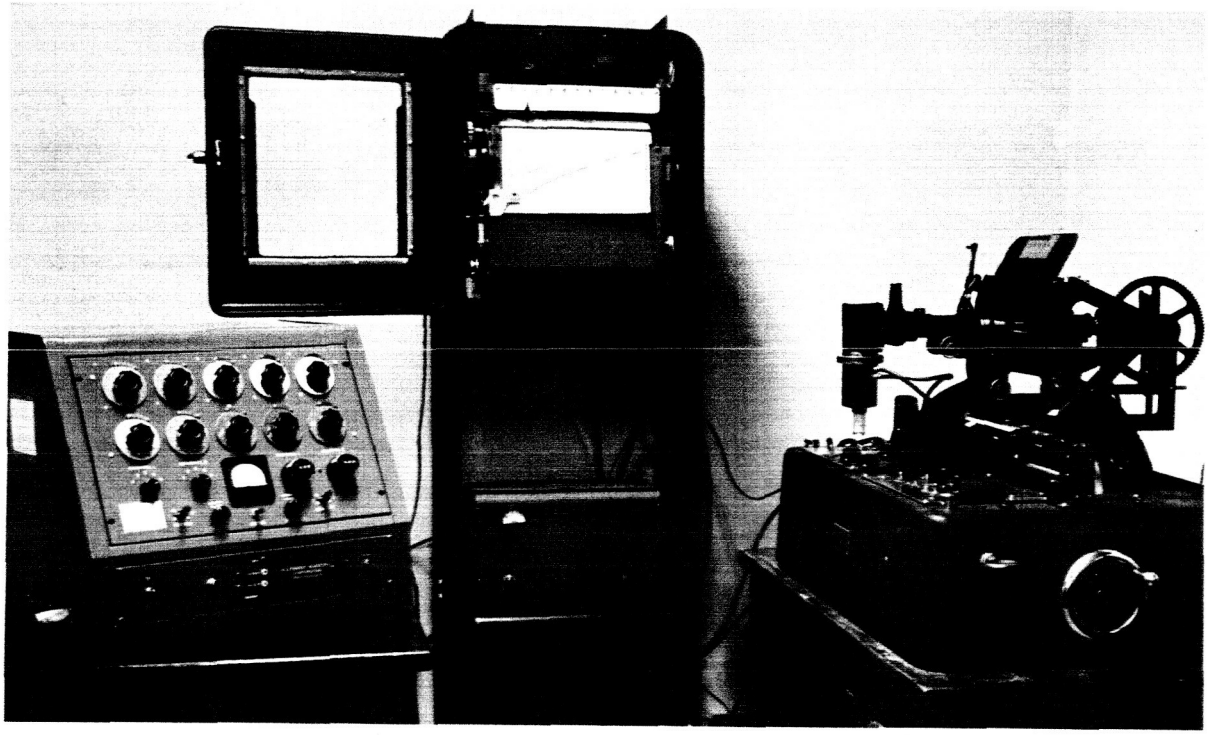

FIGURE 2.—The photodensitometric equipment used for determining the bone mass and/or density from X-ray films of living subjects. The microphotodensitometer is on the right; the recorder is central; and the analogue computer is to the left.

The small size of the rat femur amplifies certain errors, particularly those concerned with linear measurements. For example, the length of the trace path as represented on the chart paper can vary by as much as 5% because of human error in deciding its limits. This error would be reduced as the trace path length increased.

In selecting the trace path, it is essential that the bone mass per unit area across the slit be essentially constant. If the bone mass per unit area at one end of the slit varied greatly from that at the other end, the result would be an incorrect equivalent wedge thickness. The erroneous value results from giving equal weight to the distribution of intensities over the slit and comparing this average intensity with a point of equal intensity on the wedge trace. Since the response of X-ray film to X-radiation is not linear, the image resulting from the different amounts of bone mineral is not a linear function of its mass and is a possible source of error.

The method of calculating the bone density coefficient (ρ') depends on the correction for the soft tissue surrounding the bone.

The formula is as follows:

$$\frac{\text{Counter}}{\text{number}} = \text{Calibration} \times \frac{\text{Increments}}{\text{Chart division}} \times \frac{\text{Chart speed}}{(LS)\ (PS)} \times \int_a^b x'dx$$

where

counter number = the number which appears on the dial of the potentiometer

calibration = number of counts per chart increment per minute. This is an equipment function and directly influences the total count

increments = a unit on the vertical axis of the chart paper which depends on the speed with which the wedge was traced and the length of wedge

required to cover the range of film opacity
chart division = major division on the chart paper in the vertical axis
chart speed = speed of recorder in divisions per minute
LS = speed of scan of wedge
PS = speed of scan of bone image
x = distance in centimeters along the object trace path
x' = distance in centimeters along the wedge that will give the equivalent wedge thickness
$(b-a)$ = the length of the trace path (bone diameter)

The factors outside the integral are mechanical variables which are independent. These factors can be reduced to a simpler expression so that

$$\text{Counter number} = \frac{(\text{Calibration}) \times (0.1)}{(PS)(t)} \int_a^b x' dx$$

Reference to figure 3 will further illustrate the calculation procedure. It is a reproduction of an absorption trace across the wedge and across a rat femur. D is the pen deflection and x' is the distance along the wedge. The interval between (a) and (b), where $(b-a=d)$ is the femur width and also includes the pen deflection due to the soft tissue ventral and dorsal to the bone. The region to both sides represents the pen deflection due to soft tissue only. The assumption is that the deflection due to the soft tissue in the bone region can be approximated by interpolating between the soft tissue deflection on either side of the bone and soft tissue region with a straight line. The equation of this line can be written

$$D(x) = \alpha x + \beta$$

where α and β are the slope and intercept, respectively, of the line. To perform the integral

$$\int_a^b x' dx$$

x' is determined as a function of x by requiring that

$$D(x) = D(x').$$

This is automatically performed by the electronic computer. Since the wedge trace is approximately a straight line in the deflection range of the interpolated soft tissue, it may be described by the equation

$$D(x') = \alpha' x' + \beta'.$$

By substitution and performing the integral an expression is arrived at for the soft tissue counter number in the bone region,

$$\text{Soft tissue count} = \frac{(\text{Calibration})(0.1)}{(PS)(t)} dX$$

The counter number for the bone becomes

$$\text{Bone count} = \text{Total count} - \frac{(\text{Calibration})(0.1)}{(PS)(t)} k\, dX$$

The (k) is an empirically determined constant to adjust the final value to a realistic one and may be omitted if one is interested only in comparative values. The bone density coefficient ρ' is obtained from the expression

$$\rho' = \frac{(\text{Total count})(t)(PS)}{(\text{Calibration})(d^2)} - \frac{kX}{d}$$

It follows that the bone mass coefficient (BMC) can be found from the expression $\text{BMC} = \rho' d^2$.

The procedure developed for the rat femur is a general one and can be applied to other skeletal members in other animals where there is relatively little soft tissue. The method used for the phalanx was developed earlier and differs in certain details dealing with the soft tissue correction. The rat technique, applied to the phalanx, gives essentially the same results as the phalanx technique.

SOURCES OF ERROR

The radiographic, measuring, and computing procedures are subject to certain errors, some of which are unavoidable and most of which can be minimized by close adherence to prescribed operating instructions.

All the pitfalls of the radiographic procedure, from positioning the subject to final develop-

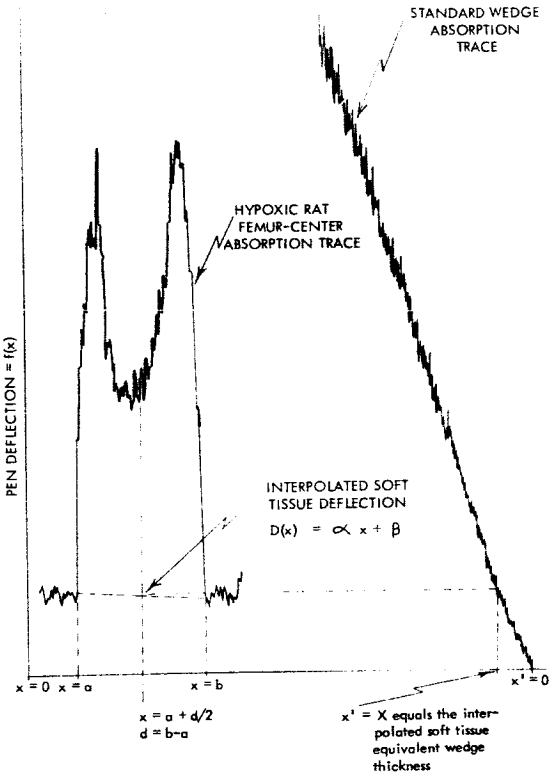

FIGURE 3.—Diagram of a densitometer absorption trace across the central portion of the femur of a living rat and along the standard wedge.

ment of the film, are prime sources of erroneous evaluations. The anatomic part to be studied must be reproducibly positioned in relation to the X-ray tube and table so that all succeeding films exhibit exactly the same trace path. The wedge must occupy the same relative position on all films. Rigid attention to these details tends to systematize errors such as the nonuniformity of the X-ray intensity over the wave front which is emitted from the focal spot of the tube.

The problem of scattered radiation can be considerable, especially when dealing with bones embedded in a large amount of flesh such as the upper femur or vertebrae. Studies in our laboratory indicate that the scattering error in the radiographic technique increases with the size of the bone and probably does not exceed 5% for most bones. The soft tissue surrounding the bone contributes the major portion of scattered radiation (Mayer et al., 1960).

Errors in measuring arise largely from failure to employ the identical trace path in repeated tracings and rarely to factors inherent in the densitometric equipment. The difficulty of locating identical trace paths can be minimized by repeating the measurements at least twice and using the average value. Also, averaging the values of several trace paths for each bone reduced this error.

The uncertainty of estimating the cross-sectional shape for computing the X-ray density coefficient introduces an error. Many irregularities in the shape of the bone cannot be observed in the roentgenogram, and the error thus introduced is virtually impossible to eliminate. Although this type of error may tend to be uniformly present in any given trace path, the efforts of age, osseous disease, and use and disuse of the cross-sectional shape have not been well characterized. Over a relatively short period of time, however, comparing the same trace path in the same individual should not seriously affect the usefulness of the technique.

In general, for intrasubject comparisons, the X-ray mass coefficient, which does not necessitate cross-section area measurement, may be more useful than density coefficients since the former removes a degree of uncertainty in the measurement.

REPRODUCIBILITY

A system of checks has been devised to test the operation of the densitometric equipment. Each day of operation, the computer is calibrated by determining the number which appears on the counter dial in one minute (counts per minute) of wide-open operation. For 100 days of operation, the mean count per minute was 562 with a standard deviation of 3.3 (an insignificant error). Another equipment check was devised as follows: An X-ray film of a wedge was traced along its length in the usual manner. The curve was then set in the computer potentiometers and the wedge image was traced again. At prescribed distances along the wedge, the corresponding counter value was taken. This reading was divided by the calibration. This procedure was repeated 30 times in order to set the limits of variation which could

be expected. The results expressed as counts per calibration for each segment of the wedge are shown in table I. The first two wedge segments are 0.5 cm apart; the others are 1 cm apart. When any succeeding wedge test film is run, always X-raying the same wedge, it is required that no more than two segment values of the wedge test film differ by one standard deviation from the values shown in the table. This procedure tests all aspects of apparatus function, including the operator. If the values do not fall into acceptable limits, the equipment is checked for malfunction. At first this procedure was followed every day; but since acceptable performance was the rule, it was reduced to every other day when the apparatus was in constant use.

TABLE I.—*Equipment Check Based on Standard Wedge*

Wedge section [a]	Section value (mean) [b]	Standard deviation
1	0.050	0.004
2	.081	.002
3	.328	.008
4	.558	.008
5	.802	.012
6	1.014	.016
7	1.267	.027
8	1.396	.026

[a] Wedge sections 1 and 2 are 0.5 cm long. All others are 1 cm long.
[b] The wedge was traced thirty times. The count for each section of wedge was divided by the machine calibration which resulted in a section value.

BETWEEN EVALUATION ERROR

A series of routine phalanx and os calcis films sent to us from a cooperating research group was used to determine the error between two evaluations of 40 films. The mean difference between the two readings was 2.66%, with a standard error of 0.40.

BETWEEN FILM ERROR

A series of subjects had two consecutive films taken of the phalanx and os calcis. Each film was evaluated twice, which is routine practice. The mean difference between the bone density values of two films for 40 pairs of routine films was 3.34%, with a standard error of 0.45. The between-film error is slightly higher than the error found between evaluation of the same film and may be attributed to differences in positioning the subject, in film processing, and other technical factors.

CORRELATION OF X-RAY MASS AND DENSITY VALUES WITH THE PHYSICAL AND CHEMICAL PROPERTIES OF BONE

Until about six years ago, density measurements were employed in many laboratories although there was little proof of the validity of such measurements from experimental data. We performed a series of experiments to demonstrate to ourselves, as well as to others, that the procedure had a sound basis as judged from empirically derived results. It was found that calibrated radiographs of excised rat femur segments and whole human femurs were highly correlated with their weights; the correlation coefficients were 0.99 and 0.98, respectively (Schraer et al., 1959; Baker and Schraer, 1958). The soft tissue problem for the rat was not a serious one since the X-ray mass and density coefficients of the intact femur and of the same excised femur were highly correlated ($r=0.95$ and 0.92, respectively). See table II for details.

TABLE II.—*Percent Change in Bone Mass Coefficient of 5-Week-Old-Rats After 24 Hours on Experimental Diets*

Group (calcium content of diet %)	N	Sum of bone mass coefficient of all trace paths		Percent change in bone mass coefficient
		Initial	24 hours	
0.025	7	0.070	0.058	−17.14
.17	6	.064	.058	−9.37
.91	7	.063	.068	+6.35
1.49	7	.060	.066	+10.00

In a more recent experiment (Schraer et al., unpublished results), an X-ray procedure was developed for experiments with the domestic hen. To demonstrate the validity of the method

with this animal, the following experiment was performed. Nineteen laying, white leghorn hens were radiographed *in vivo* for quantitative radiography without anesthesia. The tibio-tarsus seemed to be the most suitable bone from a positioning and physiological point of view. Three trace paths were chosen on the left tibio-tarsus: one was at the center of the bone and the other two were 10% of the bone length distally and proximally from the center trace path. Each film was evaluated once and a mean X-ray mass coefficient was calculated for each bone. Immediately after X-raying, the hens were sacrificed and the left tibio-tarsi were removed. The corresponding cylinders of bone delineated on X-ray images by the outer trace paths were removed, dried at 100° C, and defatted with a 1:1 alcohol-ether solution in a Soxhlet apparatus. When the weights of the dried-defatted cylinders were compared with their corresponding X-ray mass coefficients, a correlation coefficient of 0.96 was obtained (fig. 4), thus corroborating the earlier work with rats.

NUTRITIONAL AND PHYSIOLOGICAL APPLICATIONS

The efficacy of the technique has been indicated in various published studies: the skeletal response of rats to alteration of the calcium content of the diet (Schraer et al., 1963), the skeletal response of the rat to hypoxia (Hunt and Schraer, 1965), and normal and induced bone mass fluctuations in birds (Schraer and Schraer, 1961; Mueller et al., 1964).

BONE DENSITY IN HUMANS

Much data have been accumulated in which *in vivo* measurements of bone density in humans have been shown to be related to age, sex, nutritional history, and metabolic disorders. Bone density coefficients of phalanx 5–2 and the os calcis increase up to the age of 20 years and subsequently diminish in value. Females showed a more rapid decline with age than did the males (Schraer, 1958; Odland et al., 1958; Schraer, 1962). In a study of age changes in men, a loss in phalangeal density of 0.48% of age was observed over the age span 30 to 80 years (Norris et al., 1963). There appears to be a correlation between long-continued calcium intake and bone density values (Thorangkul et al., 1959); however, this relationship has not been observed consistently (Fisher and Dodds, 1958). A significant correlation in women was found to exist between both serum vitamin A and serum cholesterol and phalangeal density coefficients; a similar correlation was not noted in men. Also, a highly significant correlation

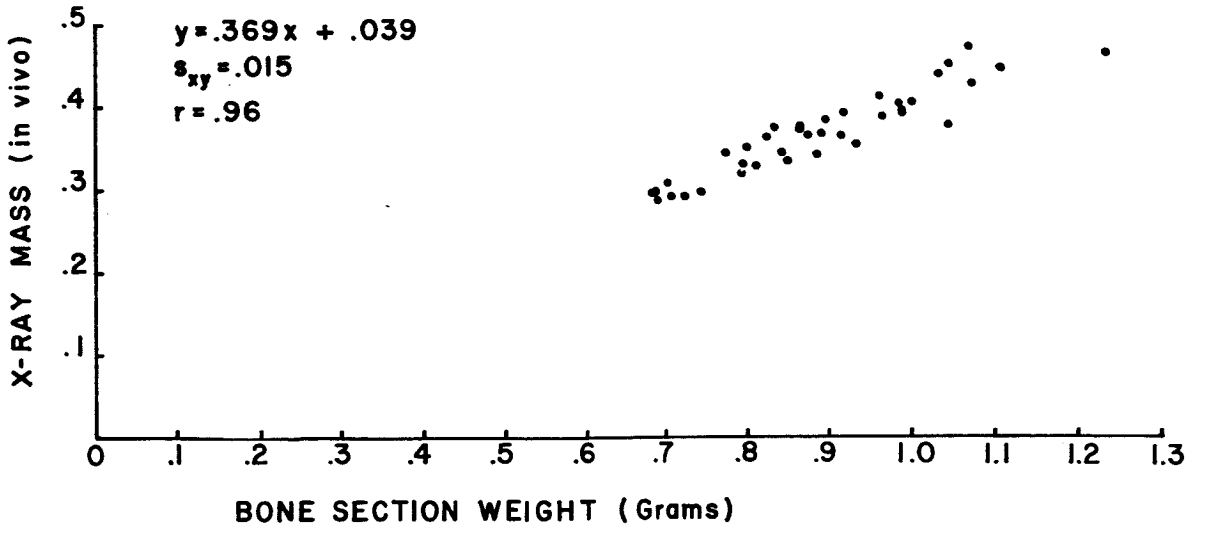

FIGURE 4.—The correlation between the X-ray mass coefficient of the left tibio-tarsus of the intact, living hen and the weight of the same bone segment removed, dried, and defatted. (Schraer et al., unpublished results.)

was observed in a geographically localized group of women between ascorbic acid intake and the os calcis density coefficient (Morgan et al., 1962).

The decreased bone density associated with age and the widespread occurrence of osteoporosis in all age groups, especially in the aged, make it important to distinguish between normal and pathological bone density values. Subjects diagnosed clinically as having osteoporosis associated with different diseases had lower phalangeal density coefficients than the average value for subjects of the same age with no known pathology (Schraer, 1962). Osteoporotic men, ranging in age from 41 to 80 years, had values which were lower than average for the same age and sex (fig. 5). In figure 6 a and b, control or average values for a particular age and sex are compared with values from subjects having different metabolic bone diseases. The lower values of the pathological subjects probably could be detected in many cases by subjective evaluation of radiography. However, the use of quantitative radiography allows one to assign a discrete number value to a particular subject with greater precision and accuracy.

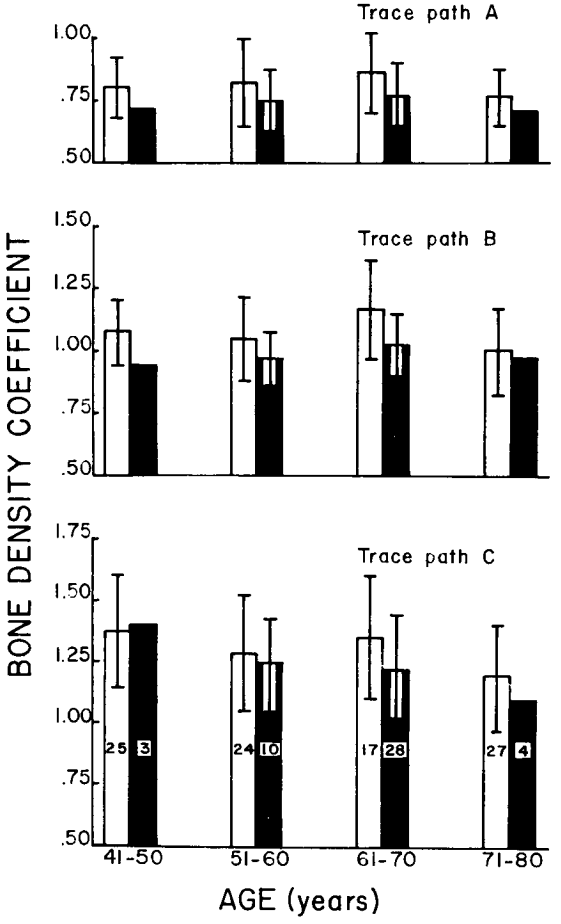

FIGURE 5.—The phalangeal density coefficients of osteoporotic men (black bars) are shown with the controls. Standard deviations are included where the numbers are sufficient for statistical treatment. Numbers of subjects for each age group are indicated on the graph of trace path C (Schraer, 1962).

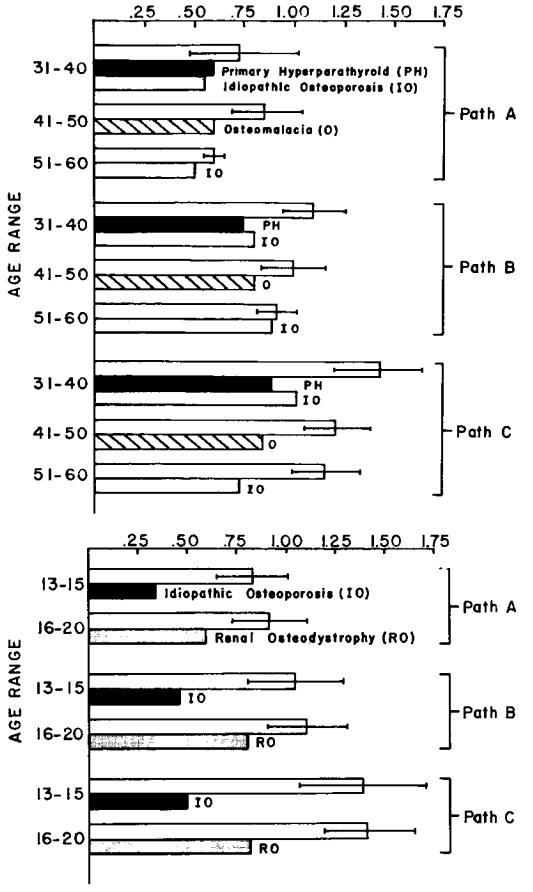

FIGURE 6.—A comparison of phalangeal density coefficients of four women (top) and two boys (bottom) with metabolic bone disease and subjects with normal or control values (white bars). Standard deviations are indicated for the controls (Schraer, 1962).

The collection of extensive data to establish average values for a particular age and sex and perhaps for a particular environment should be initiated. From these data it should be possible to establish optimal and suboptimal values and

those which are clearly pathological. These data coupled with the subjects' history may simplify and establish a firmer basis for the detection of latent pathological conditions affecting the skeletal system, for the early application of preventive medicine, and may assist particularly in the early diagnosis of osteoporosis whatever its cause.

Finally, one must be cognizant of the responsiveness of the skeletal system and the sensitivity of the technique in order to use effectively the nondestructive methods of bone mineral determination. In table III, four methods for determining bone mineral content *in vivo* are compared. For the sake of simplicity, let us assume that an individual has 1000 g of calcium in his skeleton and one is interested in determining changes in his bone mineral content as a result of treatment. The pertinent question is: How much calcium must be accrued by the skeleton before it can be detected by the same nondestructive method? Although it is well known that cancellous bone is more labile than compact bone, and that the skeletal members differ in their response to skeletal disorders, the calcium accrued in this illustration is assumed to be evenly distributed over the entire skeleton. For the hypothetical method, which has an error of 1%, it would require 100 days on a positive balance of 0.1 g calcium/day or 25 days on a 0.4-g calcium balance before detectable changes would occur if the changes were generalized. For conventional radiography, at least two years would be required on the 0.4 g day balance to observe bone changes. If these examples are reasonable, then the quantitative methods discussed here deserve wider application.

TABLE III.—*Changes in Calcium Content of a Skeleton Detectable by Nondestructive Techniques (the subject is assumed to have 1000 grams of calcium in his skeleton)*

Method of detection	% Error in technique	% Error in grams of calcium	Days required for detection depending on calcium balance	
			0.1 g/day	0.4 g/day
Hypothetical	1	10	100	25
Monochromatic radiation and quantitative radiography.	3–5	30–50	300–500	75–125
Conventional radiography	30	300	3000	750

REFERENCES

BAKER, P. T.; and SCHRAER, H.: The Estimation of Dry Skeletal Weight by Photometry of Roentgenograms. Human Biol., vol. 30, 1958, pp. 171–184.

BROWN, W. M., JR.: Bone Density Computing Machine. Proc. Natl. Electron. Conf., vol. 5, 1949, pp. 64–71.

FILES, G. W.: Medical Radiographic Technic. Second edition, Charles C. Thomas, Springfield, Illinois, 1959.

FISHER, K. H.; and DODDS, M. L.: Calcium Intake of Adolescents and Young Adults. J. Am. Diete. Assoc., vol. 34, 1958, p. 392.

HUDSON, G.: Volume and Cellular Constitution of Bone Marrow in Guinea Pigs Hypoxic from Birth. Blood, vol. 16, no. 2, 1960, pp. 1199–1204.

HUGGINS, C.; and BLACKSON, H. B.: Changes in Outlying Bone Marrow Accompanying a Local Increase of Temperature Within Physiological Limits. J. Exptl. Med., vol. 64, 1936, pp. 253–273.

HUNT, R.; and SCHRAER, H.: Skeletal Response of Rats Exposed to Reduced Barometric Pressure. Am. J. Physiol. (In press), 1965.

MAYER, E. H.; TROSTLE, H. G.; ACKERMAN, E.; SCHRAER, H.; and SITTLER, O. D.: A Scintillation Counter Technique for the X-ray Determination of Bone Mineral Content. Radiation Res., vol. 13, 1960, pp. 156–167.

MCDONALD, M. R.; and RIDDLE, D.: The Effect of Reproduction and Estrogen Administration on the Partition of Calcium, Phospherous, and Nitrogen in Pigeon Plasma. J. Biol. Chem., vol. 159, 1945, p. 445.

MORGAN, A. F.; GILUM, H. L.; GIFFORD, E. D.; and WILCOX, E. B.: Bone Density of an Aging Population. Am. J. Clin. Nutr., vol. 10, 1962, p. 337.

MUELLER, W. J.; SCHRAER, R.; and SCHRAER, H.: Calcium Metabolism and Skeletal Dynamics of Laying Pullets. J. Nutr., vol. 84, 1964, pp. 20–26.

ODLAND, L. M.; WARNICK, K. P.; and ESSELBAUGH, N. C.: Cooperative Nutritional Status Studies in the Western Region. II. Bone Density, Montana Agr. Expt. Station Bull., No. 534, Jan. 1958.

RICHTMEYER, F. K.; KENNARD, E. H.; and LAURITSEN, T.: Introduction to Modern Physics, McGraw-Hill Book Co., Inc., New York, 1955.

SCHRAER, H.; SCHRAER, R.; TROSTLE, H. G.; and D'ALFONSO, A.: The Validity of Measuring Bone Density from Roentgenograms by Means of a Bone Computing Apparatus. Arch. Biochem. Biophys., vol. 83, 1959, pp. 486–500.

SCHRAER, H.: Variation in the Density of the *os calcis* and Phalanx with Sex and Age. J. Pediat., vol. 52, 1958, p. 416.

SCHRAER, H.; and SCHRAER, R.: Bone Mass Changes in Hens observed *in vivo* During the Egg Laying Cycle. Experientia, vol. 17, 1961, p. 255.

SCHRAER, HAROLD; SIAR, W. J.; and SCHRAER, ROSEMARY: Changes in Bone Mass and Density in Living Rats During the Manipulation of Calcium Intake. Arch. Biochem. Biophys., vol. 100, 1963, pp. 393–398.

SCHRAER, H.; MUELLER, W. J.; and SCHRAER, R.: Unpublished results.

SCHRAER, H.: Unpublished results.

SIMKISS, K.: Calcium Metabolism and Avian Reproduction. Biol. Rev., vol. 36, 1961, pp. 321–367.

THORANGKUL, D.; JOHNSON, F. A.; KIME, N. S.; and CLARK, S. J.: Adaptation to a Low-Calicum Intake. J. Am. Dietet. Assoc., vol. 34, 1958, p. 392.

VAN LIERE, E. J.; and STICKNEY, J. C.: Hypoxia. University of Chicago Press, 1963.

COMMENTS

Dr. CAMERON. Here is an instrumentation question on your lens for measuring the transmission of light in your densitometer. What was the width of that slit, and how does that compare?

Dr. SCHRAER. The height of the slit is 1 mm.

Dr. CAMERON. The point I wanted to make is that if this width is comparable to your cortical thickness in these small bones, an experimental error can be introduced which will not be picked up by wedge or any other technique.

Dr. SCHRAER. That is right. In a rat, it would be very important; in a large animal, it would not be important. The scale is absolute; as you get smaller, the slit looks bigger, and I do not know the error.

Dr. CAMERON. It is true that you get better measurements of the smaller bones because of lack of tissue, and as you have larger bones, you reduce certain errors; what is the optimum of bone size using your technique?

Dr. SCHRAER. I would like to see a bone the size of the human femur that is round, with no soft tissue.

Dr. CAMERON. The term "density" is used so often here in the two contexts that it is quite confusing to me. It is film density and bone density. I think that the future speakers also should be very careful when they use the term "density" in what sense they are using this term.

Dr. SCHRAER. I agree.

Measurement of Cortical Bone Volume and Lumbar Spine Density

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Osteoporosis can best be defined as a condition in which there is a reduction in the volume of bony tissue per unit volume of anatomical bone. This reduction in the volume of bony tissue may affect trabecular or cortical bone or both. Loss of cortical bone can generally be determined by morphological measurements (except in the spine, which is a special case), but loss of trabecular bone can only be detected by changes in the absorption of X-rays or other radiation by the bone—unless the scanning method of Virtama and Kallio (1961) can be applied to man.

MEASUREMENT OF CORTICAL BONE VOLUME

Methodology

Barnett and Nordin (1960) reported three morphological measurements applicable to the metacarpal, the femur, and the vertebrae (fig. 1). The first was the thickness of the cortex of the second metacarpal of the right hand at its midpoint expressed as a percentage of the total diameter. The second was a similar measurement on the PA X-ray of the femur at the thickest point of the cortex. The third was a measurement of biconcavity in which the central height of the best-centered lumbar vertebra in the lateral view was divided by its height anteriorly. All the measurements were made on the X-rays with calipers produced by Universal Indicators, Ltd. (fig. 2). It was subsequently found that biconcavity was frequently difficult to determine on plain lateral spine films owing to overlapping of surfaces (fig. 3); and, therefore, the lateral tomogram in the sagittal plane was adopted for these measurements (fig. 4). We chose to express cortical thickness as a percentage of total diameter in order to correct for absolute size because it was assumed that big metacarpals and femora would normally have proportionately thick cortices, i.e., that there was in normal persons some correlation between cortical thickness and bone diameter. Garn (personal communication) states that there is no such correlation and prefers to express his results as absolute thickness. Figure 5 which is based on 15 measurements of male metacarpals suggests that there may be a correlation between shaft and cortical thickness.

Results in Normal Individuals

Barnett and Nordin (1960) reported results in 125 normal individuals aged 20 to 80 years and suggested that the following standards of normality might be adopted:

Metacarpal index should be over 43%.
Femoral index should be over 45%.
Biconcavity index should be over 80%.

The only one of these measurements which showed any significant fall with age was the metacarpal measurement, but both it and probably the femoral index should always be related to age since thinning of cortex appears to be an almost inevitable aging process. Biconcavity does not, however, appear to develop with age in "normal" persons, and they should probably always have an index over 80%. We suggested that osteoporosis was present when the indices

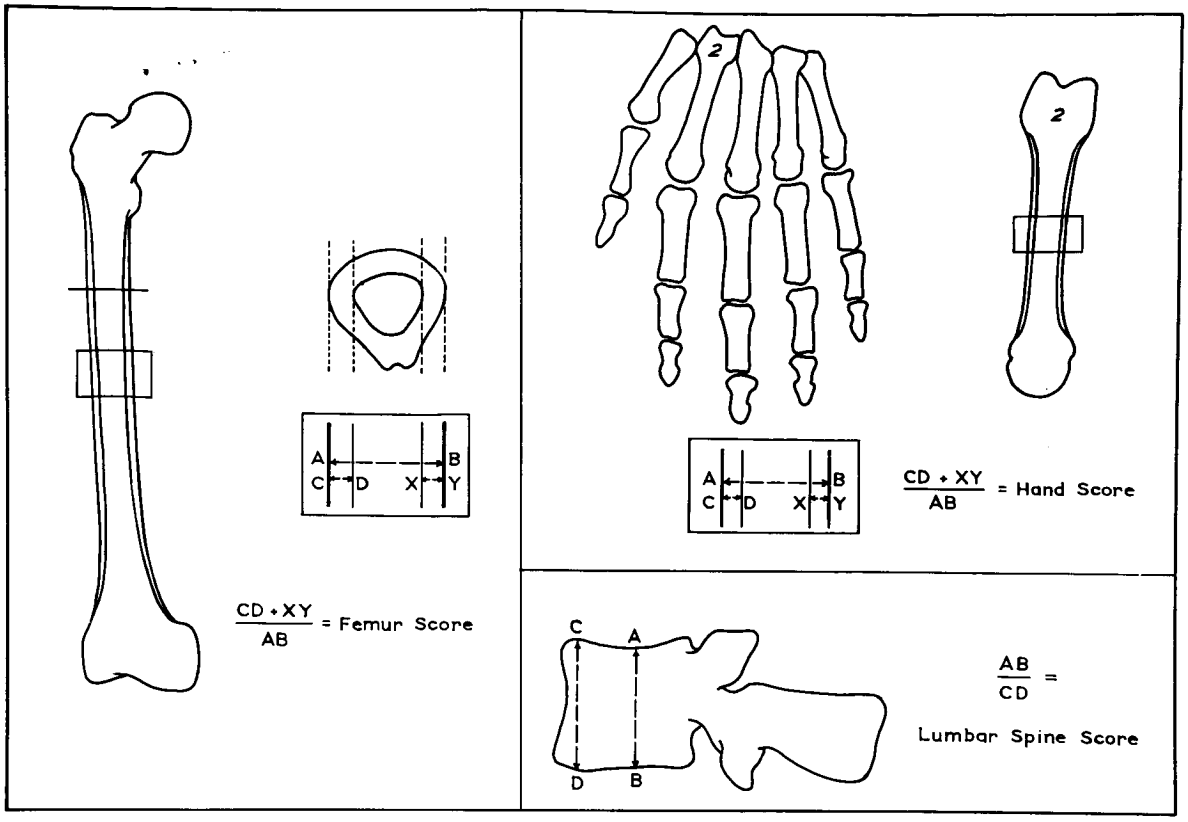

FIGURE 1.—Diagrammatic representation of anatomical measurements of metacarpal cortical thickness, femoral cortical thickness, and biconcavity.

fell below the lower normal limits and we coined the terms "central" and "peripheral" osteoporosis, the latter being present when the combined cortical indices were 88% or less.

We have applied these measurements to a study of 152 normal women in Glasgow, with the results shown in figures 6, 7, and 8. Our previous observations have been confirmed that metacarpal cortical thickness falls with age (fig. 6), but we find that this fall is very slow until the beginning of the seventh decade, when it accelerates. The only 5-year age group in which the index is significantly lower than in the preceding five years is the 60- to 64-year group. Not until the seventh decade do a significant number of normal women fall below our previous lower normal limit of 44%. Femoral cortical thickness also falls with age, but the fall is very slight until the end of the seventh decade, when it accelerates (fig. 7). Biconcavity does not appear to be an accompaniment of aging in normal women (fig. 8).

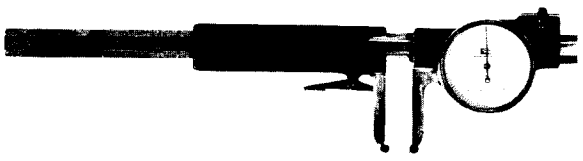

FIGURE 2.—Calipers used for X-ray measurements and made by British Indicators, Ltd.

Corresponding studies on normal men are not yet complete, but data collected so far, given in figure 9, show very little fall of metacarpal index with age. These male and female metacarpal data are very similar to those of Garn et al. (1964).

Validation of Method

Our results in clinical osteoporosis are reported in a later paper. At this stage it is sufficient to say that comparison of these X-ray

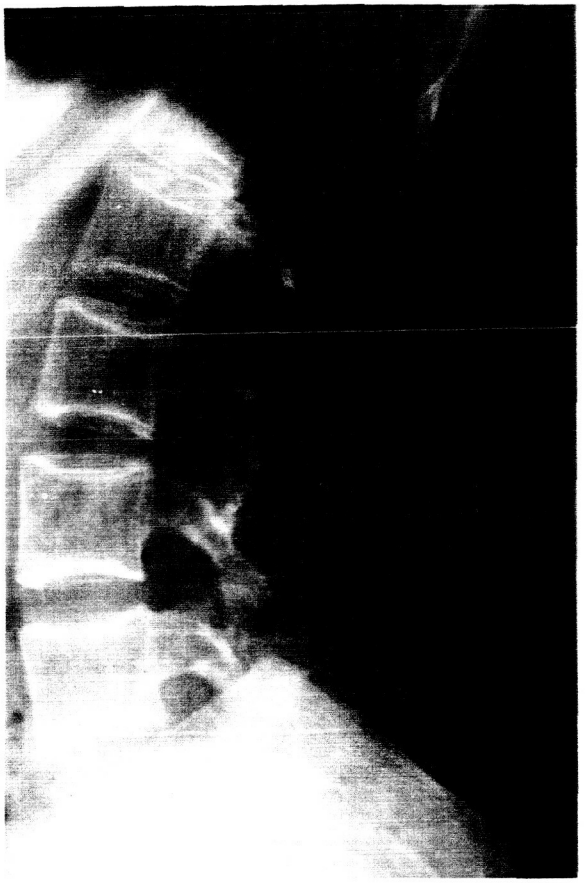

FIGURE 3.—Lateral lumbar spine X-ray in a case of osteoporosis to illustrate the difficulty in measuring biconcavity precisely.

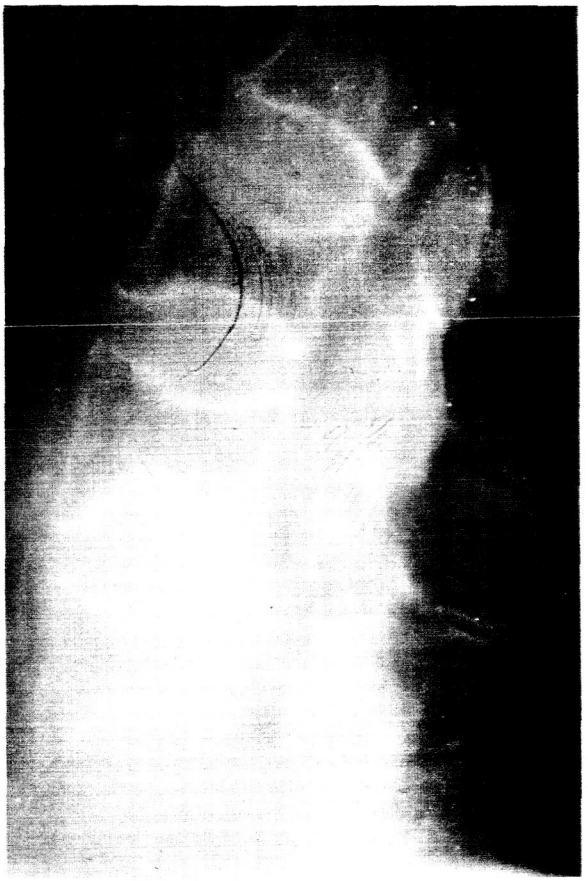

FIGURE 4.—Lateral lumbar spine tomogram to show vertebral body outline for measurement of biconcavity.

indices with iliac crest biopsies in 100 consecutive patients showed a significant correlation between the two (fig. 10). Moreover, the dividing line between the osteoporotic and normal states in the iliac crest (16% of the biopsy area occupied by the bone) corresponded reasonably well with the division between the osteoporotic and normal X-ray indices (total "score" 168/300). Nevertheless, it was apparent that these morphological measurements were relatively insensitive measures of osteoporosis, since we not infrequently saw cases of undoubted spinal osteoporosis with vertical trabeculation, obvious loss of density, or even compression, in which all the indices were normal. Thus our method was liable to false negatives though not, as far as we knew, to false positives. We therefore turned our attention to spinal densitometry.

LUMBAR SPINE DENSITOMETRY

Methodology

The main obstacle in the way of lumbar spine densitometry has always been that of correcting for the variable soft tissue mass of the subject. The basis of our method is the comparison of the density of the vertebrae with that of the intervertebral discs in order to eliminate soft tissue differences. Since this difference might be expected itself to depend to some extent upon exposure and technical factors, a phantom spine is introduced as a "standard" and the difference between disc and body density in the patient is divided by the corresponding difference in the standard. The details follow.

The patient is X-rayed in the lateral position with the phantom spine placed immediately

behind. The phantom spine consists of three lumbar vertebrae obtained from the body of a young man at postmortem. It is mounted in a perspex box, measuring 25.7 x 13.6 x 21.4 cm, filled with formol saline. The box is raised on "Temex" soft tissue equivalent rubber (supplied by James Girdler, Ltd., of London W.3) until the phantom spine is at the same level as the patient's spine. Rubber is also added at the top, if necessary, to reach the same height as the patient. A saggital plane tomogram is then taken of the patient and the phantom, using screened film with a Phillips Danatome type B. The exposure is kept, whenever pos-

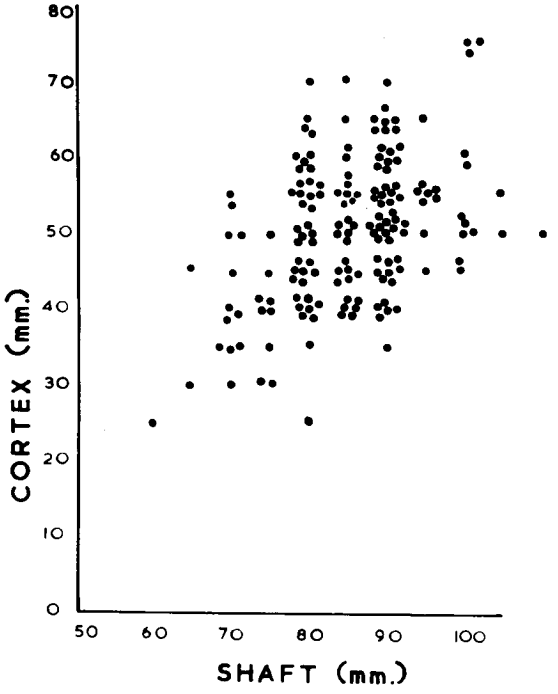

FIGURE 5.—Relation between metacarpal shaft diameter and cortical thickness in 151 consecutive observations of males of all ages.

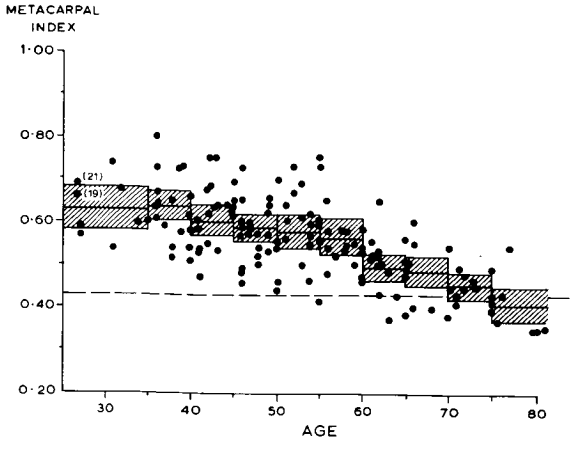

FIGURE 6.—Metacarpal cortical thickness (as percentage of shaft diameter) in 152 normal women showing two standard errors either side of the mean.

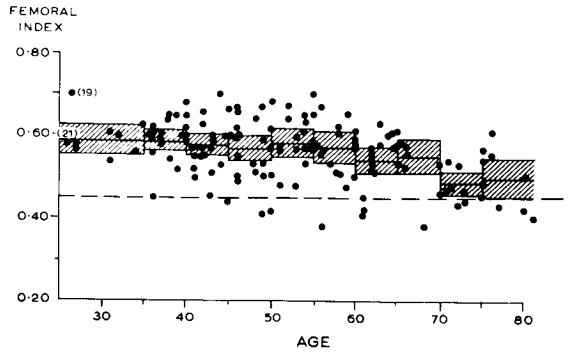

FIGURE 7.—The femoral cortical thickness (expressed as a percentage of shaft diameter) in 152 normal women showing two standard errors either side of the mean.

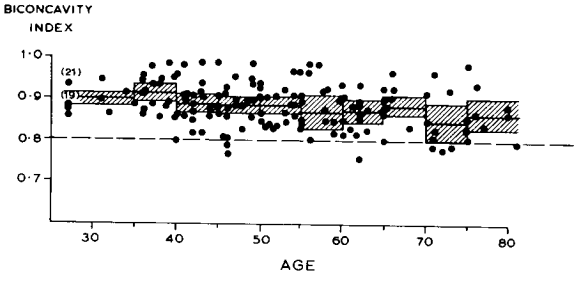

FIGURE 8.—Biconcavity index (mean and two standard errors) plotted against age in 152 normal women.

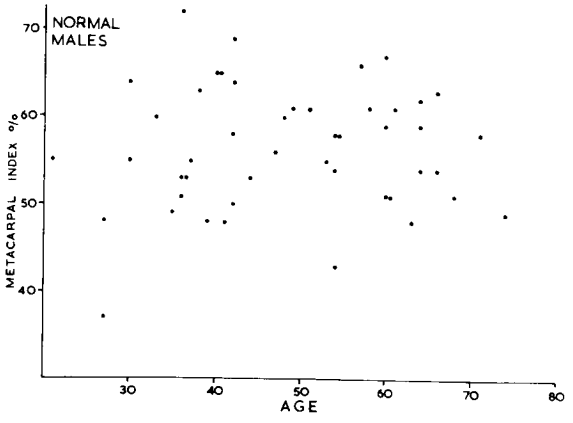

FIGURE 9.—Metecarpal indices plotted against age in normal men.

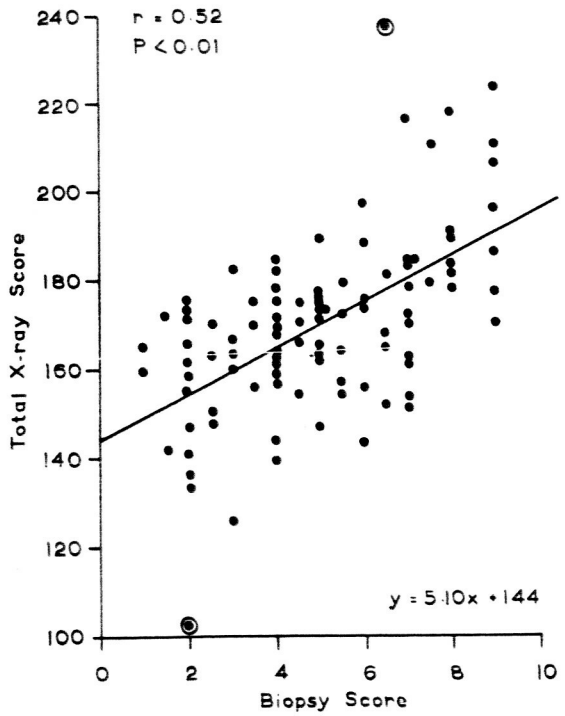

FIGURE 10.—Relation between iliac crest biopsy (scored visually on a 9 point scale) and the sum of metacarpal, femoral, and biconcavity indices in a hundred consecutive cases.

sible, within the range 80 to 90 kV, 2 kV being added per each cm of rubber. The film is processed automatically and produces an image of the type shown in figure 11.

The images of the patient and phantom spines are cut out and fed through an automatic recording densitometer (originally the Laurence Locarte, and more recently the Chromoscan made by Joyce Loebel of Newcastle-on-Tyne) to yield the type of tracing shown in figure 12. On this tracing, the end plates, vertebral bodies, and intervertebral discs can easily be distinguished. The difference between the heights of the disc and vertebral tracings is due, of course, to the bone in the latter; and the first problem is to express this difference in quantitative terms. This can be done in at least two ways. The difference can be expressed as a ratio, using either the end plates as a baseline or the absolute baseline of the instrument (100% transmission). Alternatively, the height of the vertebral tracing can be subtracted from the height of the disc tracing, and the resultant divided by the same difference on the standard tracing, which is scanned at the same densitometer setting. This is our usual calculation and is performed as follows, using the end plates as the baseline:

Average height of discs in patient (d_p) minus Average height of vertebrae in patient (v_p)----	(1)
Average height of discs in standard (d_s) minus	(2)
Relative Verebral Density (R.V.D.) = (1)/(2)--	(3)

For certain purposes we use the simple disc/body ratio, which can be expressed as dp/vp.

The heights of the disc and vertebral tracings are obtained by dividing total disc area and

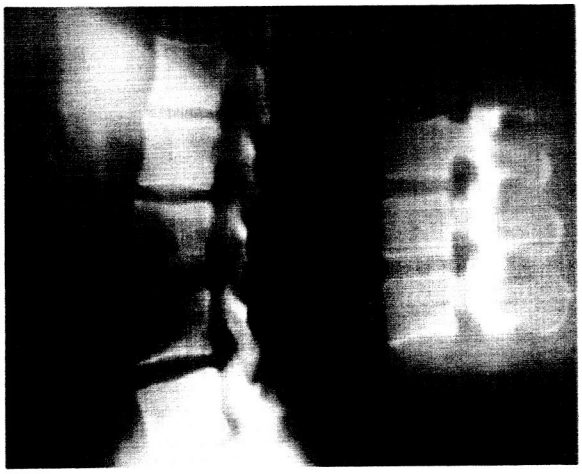

FIGURE 11.—Lateral lumbar spine tomogram on a normal subject with phantom.

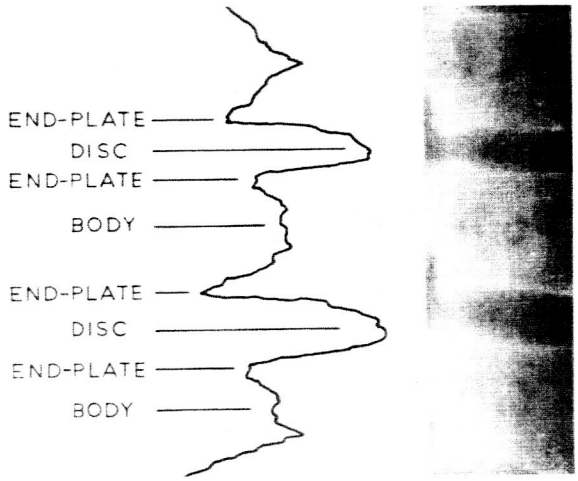

FIGURE 12.—Example of densitometry tracing obtained from the phantom spine.

total vertebral area by width at the base, i.e., between the end plates. The result is, in fact, a function of height rather than height itself; and, owing to the shape of the disc tracing, (generally triangular) compared with that of the vertebral tracing (rectangular), it tends to underestimate disc height. The original procedure (Nordin et al., 1962) was to join the peaks of the disc tracings and the tops of the vertebral tracings; but this introduced a subjective element, which should be avoided.

Effect of Exposure

Figure 13 shows the effect of exposure on the disc/body ratio of the phantom spine. The range is constant over the range of 82 to 90 kV.

Reproducibility

The reproducibility of the method is illustrated in figure 14, which shows R.V.D. values determined within one month of each other in 31 patients.

Results in Normal Subjects

The survey of 152 normal women already referred to yielded the R.V.D. values shown in figure 15. The fall with age is unmistakeable, but most of it occurs rather abruptly at about the age of 50–55 years. When menopausal age is taken into account, it is found that the fall in R.V.D. occurs in the 5–10 year period after the menopause. It is clear that it is essential to take age into account in establishing the normal R.V.D., and we would suggest that, using this method of calculation described above, it should be over 0 in women before 50 years, and over −1 in women after 50 years. The negative values must not be taken literally; they are to some extent artifactual. They arise both from the method of calculation, particularly from the division of disc height by base referred to above, and from the tomographic technique which tends to smear tissue shadows over each other, and smears the endplate shadow over the discs, making the latter appear more dense than they really are.

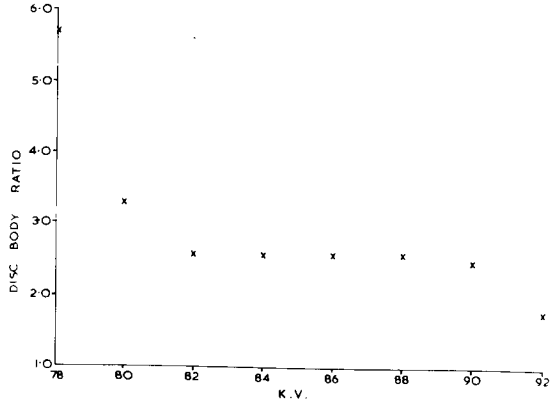

FIGURE 13.—Effect of varying kilovoltage upon the disc/body ratio of the phantom spine.

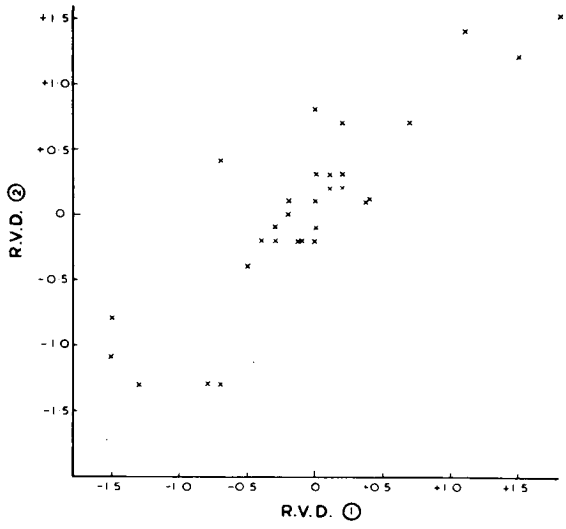

FIGURE 14.—Relation between two measurements of relative vertebral density made within one month of each other to show the reproducibility of the procedure.

Corresponding values in a small number of normal men are given in figure 16 which also shows a definite fall in R.V.D. after the age of 50.

The relation between R.V.D. and metacarpal indices in normal subjects is shown in figures 17 and 18. It is clear that the R.V.D. and metacarpal indices are related in the women but not in the men. The significance of this is not clear at present.

APPENDIX

The reproducibility of the method has been calculated from the formula

$$S = \sqrt{\frac{\sum d^2}{2n}}$$

where d is the difference between duplicate analyses and n is the number of duplicate analyses.

The results are as follows:

1. Duplicate estimations of relative vertebral density on the same film: error ±0.15 (20 films).
2. Repeat X-rays on the same patients within 24 hours: error ±0.29 (24 patients).
3. Duplicate X-rays on the same patients within one month: error ±0.31 (31 patients).

Note.—The errors given above are expressed in terms of R.V.D., the total scale of which extends from about −3.0 to +3.0. Since the error is probably not related to the R.V.D. value, it is not appropriate to express it as a percentage.

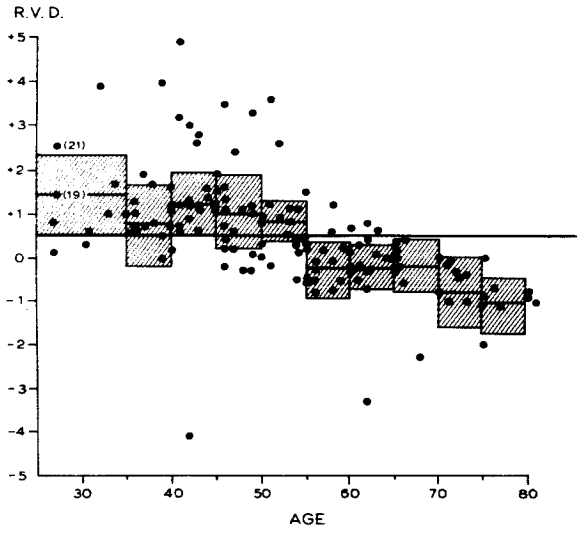

FIGURE 15.—Relation between relative vertebral density and age in 152 normal women (mean and two standard errors).

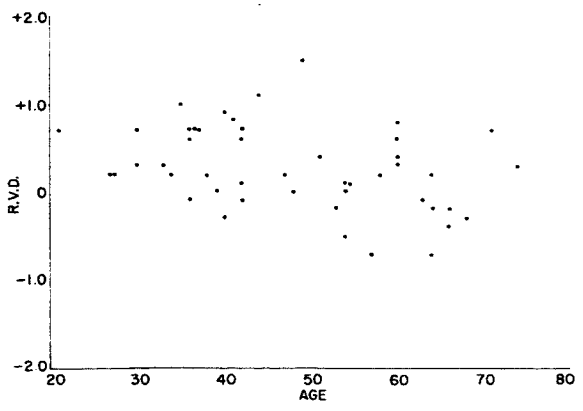

FIGURE 16.—Relation between relative vertebral density and age in 47 normal men.

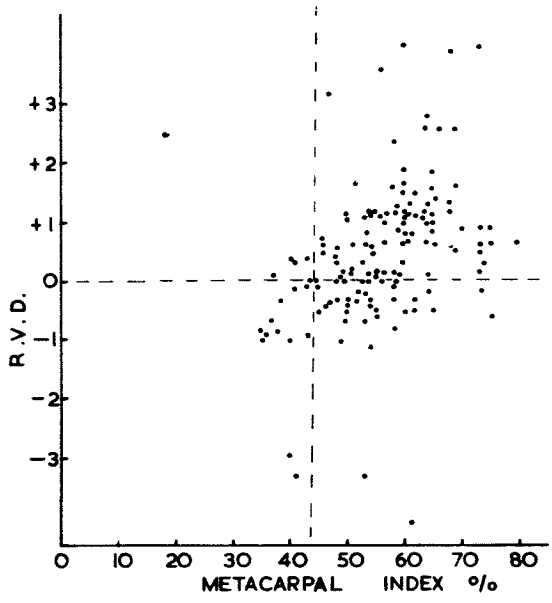

FIGURE 17.—Relation between relative vertebral density and metacarpal index in normal females.

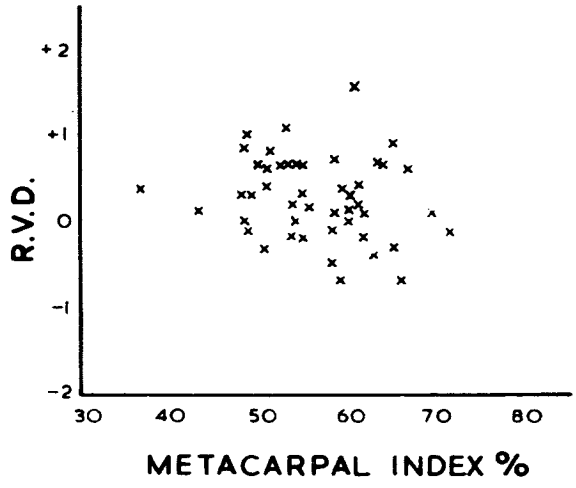

FIGURE 18.—Relation between metacarpal index and relative vertebral density in 47 normal men.

REFERENCES

BARNETT, E.; and NORDIN, B. E. C.: Radiological Diagnosis of Osteoporosis. Clin. Radiol., vol. II, 1960, p. 166.

GARN, S. M.; PAO, E. N.; and RIHL, M. E.: Compact Bone in Chinese and Japanese. Science, vol. 143, 1964, p. 1439.

NORDIN, B. E. C.; BARNETT, E.; MACGREGOR, J.; and NISBET, J.: Lumber Spine Densitometry. Brit. Med. J., vol. 1, 1962, p. 1793.

VIRTAMA, P.; and KALLIO, E.: Bone Pattern in Experimental Osteoporosis of the Rat. Ann. Med. Exp. Biol. Fennial (Helsinki), vol. 39, 1961, p. 154.

COMMENTS

Dr. SMITH. Why do you pick the anterior and not the posterior vertical height? In view of the fact that there is wedging, you may throw off your ratio.

Dr. NORDIN. That is a good question. You are absolutely right, of course. In wedging, this measurement is completely thrown out. It might be better to use the posterior vertical height as the denominator.

Dr. STRANDJORD. Which vertebrae are you using now, fourth or fifth, or which?

Dr. NORDIN. We are normally sighting on the third (lumbar), but this depends on the picture. We now do this by tomography. We started doing this on a simple lateral lumbar spine film. It works reasonably well, but there are technical reasons for technical difficulties in it. We now do it entirely by tomography on the third or fourth lumbar vertebrae, usually the third.

Dr. STRANDJORD. For the fourth and fifth lumbar vertebrae, the posterior height is frequently much lower than the anterior height, so I do not think there would be a balance.

Dr. NORDIN. Yes, that is a good point. If I were going to do it on the posterior instead of the anterior, I would have to re-evaluate the figure. It would have to be redone, but I think it is a very reasonable idea.

Dr. TROTTER. In making your caliper measurements on the films, may I ask you if you have any trouble deciding where the cancellous substance stops and the compact substance begins from the inside out?

Dr. NORDIN. This is a perfectly fair question. In pathological cases, for instance, in hyperparathyroidism, you do occasionally find, particularly in osteomalacia with secondary hyperparathyroidism and renal failure, cancellization of the inner cortex. We stated in our original paper on this that we take the point where the solid cortex ceases and the cancellization begins.

This really does not apply to these normal people since practically all of them have a sharp inner margin to their cortex, but it does apply in pathological states.

Dr. SMITH. I think this is a very important point, because the accuracy of this method is not a function of how sharp you make your point or your measurement instrument, but of where you pick the measurement point on the film, which in my experience is often 1 mm of blurred image.

Dr. TROTTER. Even in normal individuals it is not how accurate your pointer is, but what your human eye is doing, what the film technique is, I think, that is fundamental to radiogrammetry.

Dr. NORDIN. This is not a point, frankly, that has worried us in the least. In a normal case, I do not think there is any doubt where this thick cortex ends. You can see it on anybody's tracing, if you put these bones through a densitometer in the way Dr. Schraer and others have done, you can see this peak for the marrow cavity. It starts abruptly. It goes up to a point and it comes down to the other side, and you can measure it on a tracing.

Dr. TROTTER. It is about 5 or 6% error and is probably not any better or worse than what we heard earlier. It is at least that much error.

Dr. NORDIN. Yes, I should think it might well be 5% but no one is going to start drawing conclusions from 5% changes in these measurements. The changes we see are far bigger figures than we are concerned with here. When we talk about a fall in metacarpal cortical thickness with age, we are talking about differences of 50%.

The fact that our lines can be superimposed on Dr. Garn's is an indication that this is an extraordinarily simple and precise method.

Dr. ROCKOFF. I believe that if you take a large enough series or two groups and get cross sections, you will get lines that superimpose, but that is not the question we are talking about. I think the question is precision. I have used a scaled down approach to densitometry as an attempt at a tool for mensuration. It is exceedingly difficult. I think the comment is pertinent about how wide the area of uncertainty is on a radiograph, whether it is the eye or a densitometer. I think that this variance is wide.

Dr. NORDIN. I honestly do not agree with you. Of all the measurements that are fairly simple, measuring cortical thickness is one. I am simply not prepared to accept the idea that this is as difficult as you think. I am quite prepared to accept a 5% error; let's leave it at that.

Dr. RICH. It seems to me that your tomography technique will obscure, might make it hard for you to

visualize, loops of bowel with gas. On the other hand, they still would contribute something to the density.

Dr. NORDIN. Yes, you are absolutely right. One of the biggest technical difficulties is gas bubbles, and one of the advantages of tomography is that it smears the gas bubbles over the whole situation—discs and bodies; by the nature of the technique, it smears it over. I agree. We have not solved the problem of getting rid of patients' gas bubbles. If anyone can tell me how to do it, I will be most grateful.

Dr. LEROY. What interested me most was your contrast on sequential examinations of your relative vertebral densities. Can you tell us roughly what the greatest contrast was in normal adults between sequential examination, and then would you speculate on why there was a difference?

Dr. NORDIN. Do you mean what is the error? The examinations are made within a month, and I think within a month they must be called error. They very greatest difference is quite substantial. May I remind you of the method of calculation, which gives a scale of zero, where the vertebrae and intervertebral discs are of apparently equal density? If these heights are equal, the result we get is zero.

I want to make it quite clear that I think this theory of value is an artifact. I do not believe that this means that the zero on our figure means our discs and bones are, in fact, of equal density. It is an artifact of tomography, and it is an artifact of calculation. The artifact of tomography arises from the fact that you tend to smear the end plate more over the intervertebral disc, which is narrower, than you do over the body, which is wider. You have a certain spreading of density of this end plate, and it affects the disc more than the body. That tends to equalize their apparent densities. The second thing is that the disc tends to be triangular in shape, if I again may idealize; the body tends to be rectangular. If you measure your height by counting all the squares and dividing by the width, then you are unfairly reducing the height of the disc relative to the body.

There are two reasons why we tend to get a lot of negative values. I do not think this is of any importance, because our technique is essentially an empirical one, and it is essentially a relationship of one thing to another.

We are not trying to measure absolute density, and I want to make it clear that a zero value merely means that the standard and the spine are of the same relative density.

Dr. ROCKOFF. I wonder if there is not some other artifact in the technique, from this standpoint. The reason that the intervertebral disc, for instance, gets larger and the vertebral body gets smaller is that the intervertebral disc is exerting an outward force on the vertebral body. I am sure that there is a distribution of the forces that an intervertebral disc is capable of exerting. When a vertebral body collapses, while it might have had a certain optical density as it collapsed, this will appear within a short segment as if there is more calcium present. I wonder if there are not some other mechanical artifacts.

Dr. NORDIN. As far as your first point is concerned, I should have added that one of the reasons that we chose this method of calculation is because it brings out the process that takes place in osteoporosis.

The intervertebral discs expand into the vertebral body. The vertebral bodies get narrower, the intervertebral discs get wider, the relating heights change. Therefore, if you do your calculations by dividing the number of squares by this width, you have a calculation which brings out the osteoporotic processes, both from the densitometric point of view and from the morphological point of view. This does not apply to any of my normal data, because that normal series does not include any cases regarded as pathological biconcavity.

With regard to compression fractures, I agree with this entirely. Whereas a traumatic fracture will give you an increase in the density of the vertebral body which is fractured, an osteoporotic fracture collapses because there is a loss of bone within it. Therefore, although you may not get such a low density as you would expect in that vertebral body if it was not collapsed, you would not expect to get an increased density, because it is collapsing because of loss of bone.

Dr. SMITH. I think we may be in error in assuming that when you have fish-mouthing and biconcavity that there is a density increase which is just due to, let's say, microcompression. There is some evidence that this may be an internal remodeling in the vertebral body at a constant density in adjusting to the forces exerted by the intervertebral disc; therefore the density might not necessarily increase with biconcavity.

Dr. NORDIN. This could be one of the reasons why most cases with biconcavity do not necessarily have as low a density as you expect. I do not want you to think all the patients with biconcavity have a normal density. There are interesting exceptions with biconcavity and with quite normal, and occasionally even high, density.

Dr. ROCKOFF. I think it is apparent that by subtracting the way you are doing, you are assuming that the relationship between the density on the film and the calcium or the mineral content is a linear one. You are assuming that X amount of calcium is lost, Y amount of change in density is occurring, and this relative relationship will apply no matter where you are on the calcium loss curve.

Dr. NORDIN. Yes, I think you would be absolutely right in saying we were assuming that, if I had tried to express something in milligrams of calcium, but I have not.

Dr. ROCKOFF. We are assuming it by subtraction.

Dr. WHEDON. You have the same implication mathematically, though, whether you give it a unit measurement or not.

Dr. NORDIN. What you are saying is that the position of this in terms of the meaning of this subtraction will be given on different points of the scale.

Dr. WHEDON. Yes. That is exactly it.

Dr. NORDIN. I must agree with you entirely on this, and we are trying at the moment to devise a method with an aluminum standard that would get around it. If we keep within that kilovoltage range where the disc body ratio is constant, we are reasonably safe, but I am sure in precise terms that you are right.

Dr. GARN. The Joyce Loebel response is not quite log linear with respect to a linear wedge, so that the high peak is much more than additively higher than a lower peak.

Dr. NORDIN. Although I am now using the chromoscan from Joyce Loebel, the work on which this paper was based was done on a Lawrence Lockhart densitometer, which converts from a log function into a linear function. You have the same difference, wherever you are on an absolute scale, on the Lawrence Lockhart.

This is not true of the Joyce Loebel machine. The position where you are in absolute density affects the difference on the Joyce Loebel. We are trying now to introduce something like an aluminum standard to take care of that.

Dr. WHEDON. I have a different question. Why have people selected L–3 as the choice vertebrae to examine? In my clinical experience, which is not extensive, I see more fractures occur along the spine from about thoracic-8 to lumbar-2 or 3, with the most frequent occurrence between T–10 to L–1. Why did you and Dr. Vose also pick L–3?

Dr. NORDIN. We were not looking for nonfracture.

Dr. WHEDON. Yes, but the area I have mentioned is where the greatest degree of change in mineral density occurs, above L–3, as evidenced by fracture.

Dr. NORDIN. We were entirely concerned with getting the vertebral body that was best centered for a measure of biconcavity. With tomography, it does not really matter, but before tomography it mattered a tremendous amount. If you want to be right in the middle of this beam, and if you take a picture on the lumbar spine, L–3 will normally be in the middle.

Now, you can ask the radiographer to start taking pictures at the thoracolumbar junction, but the normal lumbar spine picture will be centered at L–3, and that is the reason we chose it.

Dr. STRANDJORD. Biconcavity occurs clinically far more commonly in the lumbar region, but wedging, anterior wedging, is in the thoracic.

Dr. NORDIN. That is right, because of the shape of the spine.

Dr. WHEDON. This is quite true, but I suspect that if one were to look at a large series of films, one would see more compression fractures at an area above L–3.

Dr. VOSE. We selected L–3 instead of the thoracic vertebrae because of the ribs. Overlapping ribs make it very difficult to get densitometry. In addition, we selected L–3 because it is the easiest to obtain in autopsy.

Dr. NORDIN. That is a good reason.

Radiographic Bone Densitometry

PAULINE BEERY MACK
Nelda Childers Stark Laboratory
Texas Woman's University

Beginning in 1927, various colleagues and I developed a series of instruments designed to measure bone mass and density. The evolutionary progress leading to our present instrumentation is described in the open literature.

At Texas Woman's University we reconstructed a Model IV while working on a new plan for a bone density computer, which was designed and built from 1954 to 1957 with the cooperation of physicists at the Leeds and Northrup Company, Philadelphia (primarily the late James Donald Nelson). Bone Densitometer Assembly VI was described by Nelson et al. (1958) and by Mack et al. (1959).

INSTRUMENTATION OF CURRENTLY USED BONE DENSITOMETER ASSEMBLY

Assembly VI currently used in our bone mass measurements at the Texas Woman's University is a special purpose analog computer consisting of an electromechanical servomechanism and an integrating unit. The major units of the instrumentation used in evaluating the radiographs in these laboratories are shown in figure 1. The equipment consists of five major subassemblies, all designed to operate together as a completely integrated system. The basic units of the overall assembly are the following:

(a) A modified Knorr-Albers scanning unit (unit at right of the figure);
(b) A Speedomax Model G transmitting recorder (unit in center of the figure);
(c) A series of 20 potentiometers in the same panel as (b);
(d) A Speedomax Model G recording potentiometer; and
(e) An Instron integrator.

Figure 2 consists of a schematic diagram of the integrated microdensitometer system. Following this diagram, a signal is given by the scanning unit which is received by the first recorder as an uncorrected trace of a wedge on the radiograph being scanned. An adjustable slidewire is mounted in this recorder, which serves as a function generator, tapped at 21 points to give 20 sections of equal length. Twenty potentiometers are associated with the 20 sections of the slidewire to permit manual adjustment for purposes of correcting the first wedge trace. After the function generator has been adjusted, the circuit setting is transferred to the receiving recorder, thus permitting a corrected wedge scan as well as a corrected bone scan to be received and displayed on this recorder. The bone trace gives a readout count on the integrator.

Scanning Unit

The scanning unit consists of an optical system, a plate stage, a drive mechanism for the plate stage, and a d-c amplifier for the recorder. The optical system includes a special tungsten lamp with plane optically polished windows, powered from a highly stabilized power pack having a constancy of output with 0.1% for a-c line voltages between 100–125 volts at freguencies between 55 and 65 cycles. The beam from this lamp is focused on a photocell after

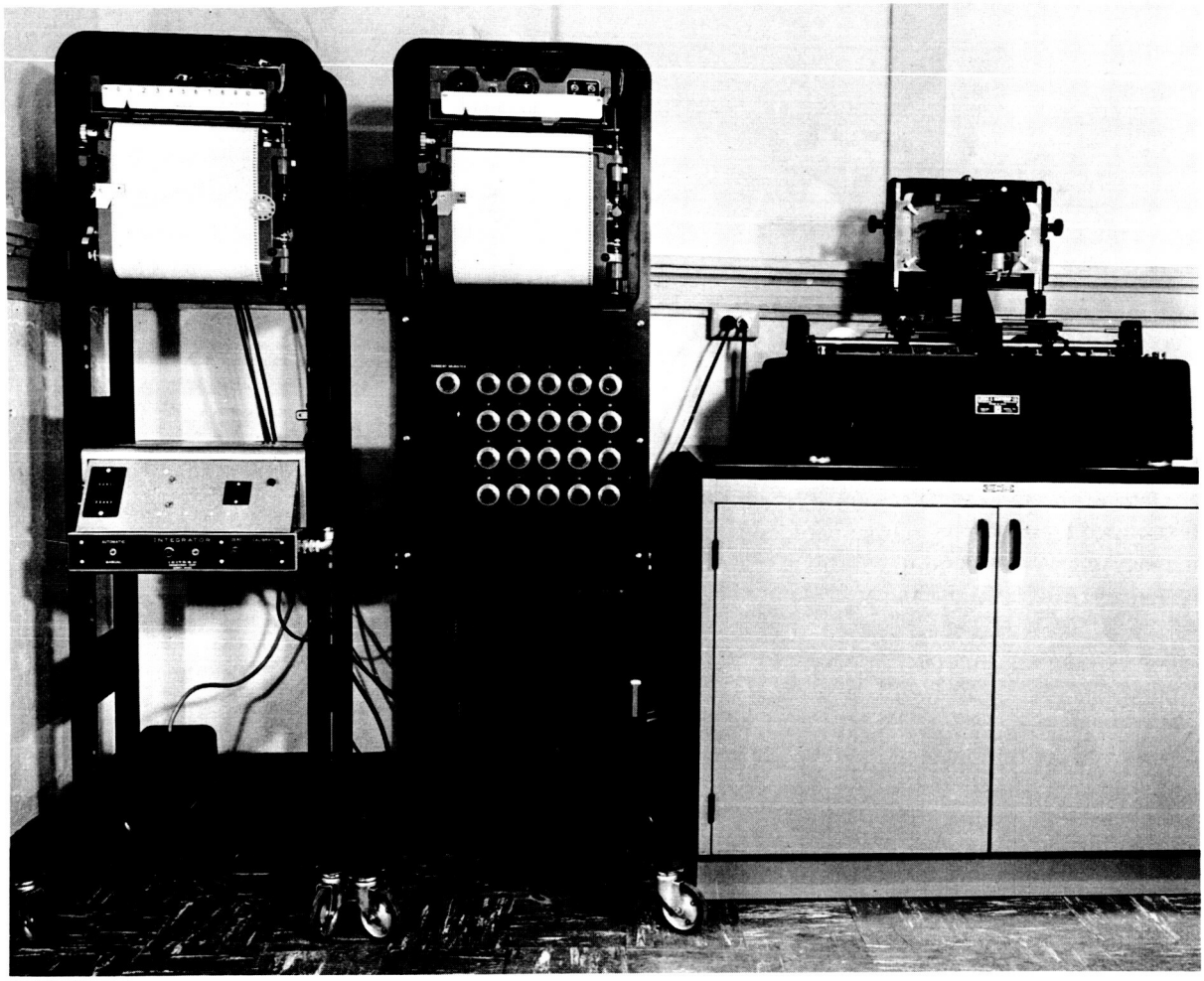

FIGURE 1.—Bone Densitometer Assembly VI currently used at the Texas Woman's University. *Right:* Densitometer. *Center:* Transmitting recorder with potentiometers for correcting calibration wedge trace in lower part of panel. *Left:* Recording potentiometer with Instron integrator in lower part of panel.

passing through the X-ray plate being scanned. The film is mounted on the plate stage that is supported by ball-bearing rollers on a carriage rod, all accurately machined to very close tolerance.

The drive mechanism for the plate stage has nine selectable speeds, varying from 0.1 mm per minute to 50 mm per minute. Each plate travel speed is regulated closely by a synchronous motor drive; and adjustable limit switches govern the limit of travel in either direction, as the plate travel is conveniently reversible by means of a switch.

A scale, mounted on the scanning unit and calibrated in millimeters, subdivided by a vernier, indicates plate travel and enables the operator to scan a number of precisely equal segments of the film trace. This scale also permits the operator to retrace *exactly* the same length of film on repeated scans and serves as the guide for integrating the equal trace segments. The plate travel is synchronized with recording chart travel to insure that quantitative measurements of density can be produced and reproduced accurately. A precision d-c amplifier with stabilized zero and stabilized gain multiplies the minute current from the photocell to a value measurable by a self-balancing potentiometer recorder. A switch on the scanning unit increases the amplification fivefold when needed.

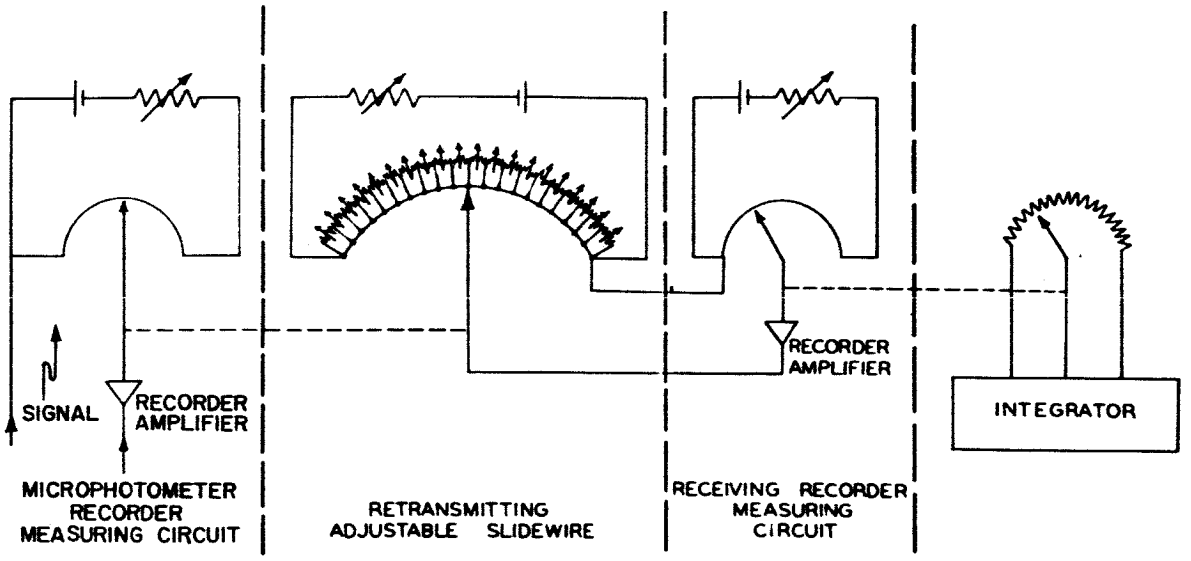

FIGURE 2.—Schematic diagram of the integrated densitometric system illustrated in figure 1.

First Recorder

The first recorder (center unit in fig. 1) consists of a Speedomax Type G self-balancing functioning recorder having adjustable zero, adjustable span, and a full scale balancing speed of less than one second. This recorder indicates continuously the magnitude of the amplified photocell current and traces a graph on its chart in synchronism with the scanning unit plate travel.

A major feature of the complete assembly consists of a special d-c retransmitting slidewire mounted in this Speedomax self-balancing potentiometer recorder, with a moving contact on this slidewire driven in synchronism with the recording pen in an indicating pointer. This retransmitting slidewire is divided very precisely into 20 equal segments, each segment being shunted by an adjustable ten-turn potentiometer. An adjustable d-c voltage is impressed across the total slidewire; and the output of the slidewire is characterized by adjusting the 20 potentiometer dials to provide a calibrated output from this slidewire from the trace of the reference wedge.

The uncorrected wedge trace on the first recorder is scaled by the operator at 20 equal intervals, using a special transparent rule calibrated to provide direct setability of a calibration factor on the one thousand division dials of the 20 potentiometers.

Since the scale of the first recorder is determined by the percentage of transmitted light and the calibrating retransmitting slidewire corrects the scale of the second recorder to standard wedge density, producing a straight line trace of the calibrating wedge in conformity with the wedge slope, the X-ray absorption as indicated by its densitometer trace on the second recorder is related directly to the X-ray absorption of the standard wedge.

Second Recorder

The second recorder receives, displays, and records the signal from the special calibrating retransmitting slidewire which serves as a function transformer, providing a line graph of the reference wedge and a calibrated density trace of the bone sample. Its graph also is synchronized with the automatic scanning. A retransmitting potentiometer is driven from the output shaft of this second recorder and actuates an Instron integrator which is located in the base of this unit.

Instron Integrator

The integrator provides a digital readout proportional to the area under the calibrated den-

sitometer trace of the bone section which has been scanned on the second recorder. The integrator is equipped with two digital counter readouts. One is reset after each segment is integrated, if a sectional trace is segmented, while the second readout acts as a continuous counter, providing a totalized check on the readings taken from the first counts. A bell actuated by the limit switch on the plate travel mechanism signals the operator that the desired scan travel has been completed, and the integrator counting is stopped at this point.

SEQUENCE OF OPERATIONS IN MAKING A BONE MASS DETERMINATION

To obtain a satisfactory calibration for a trace of a bone section from an X-ray film, the fundamental step is provided in the use of a calibration wedge on the same film as the bone image, as shown in figure 3. The wedge is made of an aluminum alloy, as noted, with X-ray absorption characteristics similar to that of bone. The wedge used on an os calcis trace, which is traced from its beginning to a distance of 13 cm along the base, has a slope of 1:10 and thus provides a linear calibration unit from 0 to 1.3 cm of aluminum alloy wedge equivalency. The range of film density for an os calcis in normal adult males corresponds to the film density along 50 to 80% of the wedge length.

The initial record obtained on the first recorder from scanning the wedge with the densitometer without correction provides the basis for calibrating the wedge curve for the film. Represented in this tracing is the percent light transmission through the film for displacement along the wedge from the beginning to 13 cm out on the wedge trace. The shape of this uncorrected curve is influenced by many factors inherent in the film itself, the film exposure, and the film processing technique.

It is desirable to integrate the equivalent area under a tracing obtained across a line on a bone X-ray to provide an expression of equivalent bone mass for a given film. Since the roentgenographic process distorts a linear graph which would represent the wedge accurately, the units of light transmission must be calibrated to equivalent wedge thickness before integration of the area under the tracing of a bone, if the results of the latter are to be correct This calibration procedure is provided through an electromechanical servomechanism which uses the wedge trace to provide calibration factors over 20 segments of the wedge trace, thus providing accuracy in the latter and the basis for accuracy in the subsequent bone trace.

FIGURE 3.—Radiograph showing lateral view of human adult male subject, with the image of the os calcis calibrating wedge on the radiograph. The line represents "conventional" posterio-anterior trace made for purpose of measuring bone mass in this central section.

Note.—Other parallel traces may be made the distance apart of the Densitometer scanning beam above and/or below the trace which is illustrated, in order to measure the bone mass of a wide band in this bone.

The sequence of operations needed to achieve calibration of a density curve and to integrate the area under the curve of a bone on the same radiograph follows.

(a) The wedge roentgenographic image first is scanned for the purpose of providing the density calibration curve of that film on the first recorder. Before this, the first recorder is balanced to zero for the "no light transmission" condition obtained by inserting an opaque ob-

ject between the light source and the photo cell. Full scale output of the recorder is obtained for the light transmission 13 cm out on the wedge trace by adjusting the light source voltage and/or the amplifier gain.

Once this adjustment is made, it remains the same throughout the rest of the procedure. In this manner, the extreme limits of the wedge are set the same for each procedure. The use of a site on the wedge other than 13 cm out the base would require corresponding changes in the multiplication factor for converting integrator units to equivalent wedge mass. The uncorrected curve for the wedge then is traced on the first recorder.

(b) Then the technologist measures the curve at 20 points along the chart and sets the 20 potentiometers in the panel associated, respectively, with each of the points to provide a calibration correction factor for each of the 20 divisions. The current through the slidewire and its shunted potentiometers then is set to give full scale deflection of the second recorder for the zero light transmission for transmission 13 cm out the wedge.

The retransmitting slidewire is mechanically coupled to the first recorder tracing which is proportional to the electrical inputs. In this manner, a calibration factor is applied by each potentiometer according to the reading of the first recorder, so that the output can be used to drive the second recorder to display a calibrated output curve.

(c) When the potentiometers are given the settings required to obtain the needed resistance for calibrating the original wedge trace, a retrace of the wedge is made on the second recorder. If the potentiometers have been adjusted correctly, a uniformly straight graph is obtained.

(d) After the second recording potentiometer has recorded the signal from the special transmitting slidewire and has received and displayed the corrected wedge trace, a calibrated density trace is made of a section of a bone on the same film as the calibration wedge image. This bone trace, also displayed on the second recorder panel, is synchronized with the automatic scanning system.

Since the scale of the first recorder is linear with the percentage of transmitted light and since the calibration procedure has corrected the scale of the second recorder to standard wedge density, the X-ray absorption by the bone section as indicated by its densitometer trace is related directly to the X-ray absorption by the standard wedge.

(e) A retransmitting potentiometer is driven mechanically from the output shaft of this second recorder which actuates the Instron electronic integrator, providing a digital readout proportional to the area under the calibrated densitometer bone section trace, as noted.

CONVERSION OF RESULTS IN TERMS OF INTEGRATOR COUNTS TO OTHER UNITS

The counts secured from the integrator may be used directly from one bone mass evaluation to another, since a conversion of integrator counts to another factor such as equivalent wedge volume or equivalent wedge mass would involve the use of the same conversion factor for each set of counts, thus resulting in the same percentage change from a radiograph of one subject made at one time to that made at subsequent times. On the other hand, the counts can be converted readily by a mathematical formula to equivalent wedge volume and thence to equivalent wedge mass, the latter because the density of the aluminum alloy wedge is known.

The aluminum alloy wedge has been used to calibrate calcium compounds which are components of, or are associated with, bone mineral. The calibrations have been made by placing chemically pure calcium compounds, or mixtures of compounds of known weight, in plastic encasements and exposing them on the same film as the standard wedge. The wedge image and the image of the chemical compound then can be traced in their entirety with parallel crosswise scans running the length of each image.

As a result of such a calibration we sometimes report bone mass data of living subjects in terms of "calcium hydroxyapatite equivalency." The reporting of bone mass in such a term is not intended to denote that the bone

under consideration contains this amount of calcium hydroxyapatite but that the substances in the portion of the X-ray which was evaluated had an X-ray absorption value equivalent to the designated quantity of this calcium complex. The materials present in living bone and under- and over-lying tissue include bone mineral, protein, water, and fat. The respective contributions of the separate substances to the mass absorption coefficient of the entire bone tissue are dependent upon the exposure energy applied to the bone.

Figure 4 shows the relationship of the X-ray determined mass of 50 ashed bones to the mass of the ashed bones as determined analytically, which is linear. The bones ranged from those of a weanling rat to those of human subjects, as well as those of large farm animals. The bone ash in each case was traced in slices which were the thickness of the scanning beam through the entire length. The results are reported as mass of bone ash in terms of calcium hydroxyapatite, the latter having been calibrated against the alloy wedge exposed on the same film.

SELECTION OF TRACING PATHS FOR BONE MASS EVALUATIONS

Evaluation of the Os Calcis

It has been mentioned that the os calcis was the first anatomical site to be chosen during the early period when a method was being sought to evaluate bone mass through the use of X-rays and when equipment for this purpose was being developed. The next bone to be added was the second phalanx from the distal end of the fifth digit. In the reports published from 1938 throught 1942, both of these sites were used in arriving at conclusions concerning skeletal mineralization.

Somewhat later the patella was added, and these three anatomical sites have been those chiefly used in our work to date. Two very important sites have been studied by my colleague, George Vose, namely, certain lumbar vertebrae and the femur neck. He is reporting his progress with these two difficult locations separately.

Reason for Choice of Os Calcis.—The reasons for the selection of this bone were the following: (1) The os calcis is a large bone with a minimum of over- and underlying soft tissue, which has several advantages including the reduction of X-ray scattering. (2) This bone is easily accessible for X-raying. (3) This bone contains a large amount of cancellous or trabecular tissue. It is stated by Cantarow and Shepartz (1962) that interchange of ions between bone and extracellular fluid occurs more readily in newly formed trabecular bone than in older compact bone. (4) The foot is subjected to a

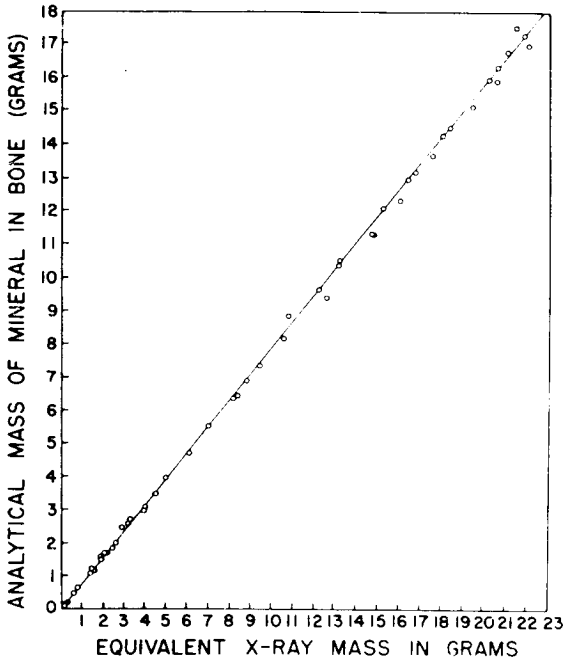

FIGURE 4.—Linear relationship of the X-ray determined mass of 50 ashed bones of a wide variety of weights to the analytically determined mass in terms of calcium hydroxyapatite. The following conversion factor was used:

$$\text{Equivalent Wedge Mass} = \frac{\text{S.L.V.H.C.}}{K} \text{ (cm}^3\text{) } D$$

Where S=Slope of wedge (cm/cm); L=Baseline length of wedge portion traced (cm); V=Velocity of scan (cm/min); H=Vertical dimension of the scanning slit (cm); K=Counts obtained for one minute of full scale integration (counts/min); C=Integrator counts; and D=Density of aluminum alloy wedge.

Note.—Hydroxyapatite powder packed tightly into a plastic encasement had a density 1.075 times that of the aluminum alloy wedge.

great deal of mechanical stress with normal activity, and therefore it is likely to show differences in bone mass in persons who differ markedly in their level of activity or in one person while active and again while immobile. (5) The os calcis in the adult has two distinct anatomical landmarks which can be located without difficulty and which serve to mark the extremes of tracing paths which are valuable both in mass studies in which various populations are compared or in longitudinal studies involving one group of humans beings evaluated repeated times.

Figure 3 shows a radiograph of a lateral view of an os calcis with the calibration wedge in place. The calibration wedge shown in this illustration consists of the aluminum alloy described above. It is machined with a 10 : 1 slope, developed specifically for this anatomical site. The line drawn in this figure shows the location of a scan which we have called our "conventional" os calcis tracing path. This represents a section of bone from a posterior to an anterior landmark 1.3 mm wide, the width of the scanning beam.

In making scans of the conventional tracing path in successive films of one subject, pricks are made with a steel needle at the extremities of this path, but outside the bone, with a new film carefully superimposed over the original film. The same superimposition of wedge film over wedge film also is made in order to insure that the same path of the calibration wedge is traced from one time to another.

In figure 5, the cross-sectional view of the os calcis through which this scan has been traced is divided into 10 segments from the posterior to the anterior end of the bone. These divisions are possible because the densitometer can be stopped at desired intervals, as mentioned in the description of the instrumentation, with the integrator counts given for each segment. This segmentation procedure enables changes in bone mass to be recorded in posterior segments of this bone where the skeletal tissues tend to be more dense, in central segments where there is a more lace-like configuration, or in the anterior segments.

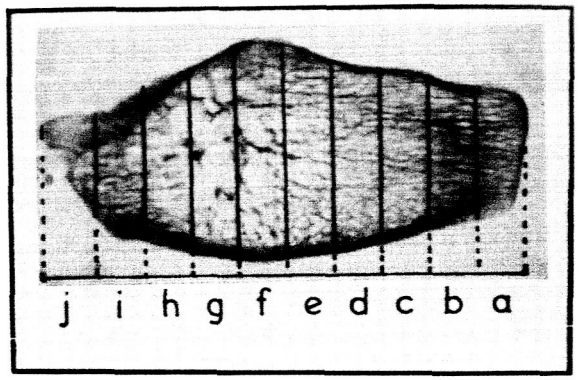

FIGURE 5.—Cross-section of os calcis through "conventional" scanning path in radiograph illustrated in figure 3.

Note.—The Densitometer is stopped by the operator at intervals to obtain integrator readings on 10 segments of the total section. This enables observations to be made of bone mass changes in the various parts of this bone section.

In order to find the bone mass of larger proportions of the os calcis, scans may be made 1.0 mm apart below and parallel to the conventional trace down to the bottom of the bone and above the conventional trace as far as possible without including any overlapping ankle bone.

No further view of the os calcis need be taken if bone mass is the factor to be measured. The entire bone, exclusive of bones which overlap the os calcis at the top, can be included in bone mass measurements by means of the parallel traces described above on the lateral view which is taken. On the other hand, a band of any width can be measured by means of parallel scans across this bone parallel to the conventional scan.

Evaluation of Finger Phalanx 5–2

The middle phalanx in the fifth digit was selected as an anatomical site for evaluation because it represents a bone which contains a substantial amount of compact skeletal tissue and because it is easily accessible for radiographing. The selection of a single phalanx in the fifth digit was made because of the finding that the bone mass values of other phalanges in the same subject were closely correlated. Nevertheless, in some longitudinal studies more

than one phalanx may be used, and a selected metacarpal as well as selected carpals may be evaluated also.

Evaluation of the Patella

The patella was selected for evaluation for bone mass because it contained both cortical and cancellous tissue and because it is easily accessible for X-raying.

Positioning of Subjects for the Three Radiographs

Subjects lie flat on their backs with a flat pillow under their heads for making exposures for the three bones—namely, os calcis, finger, and patella. They are covered entirely with leaded rubber sheeting except for the small part of the anatomy being X-rayed.

The X-ray of the hand is taken while the subject is flat on his back. Then he is turned slightly to his left to position the left os calcis and still further left to position the left patella.

All three radiographs are exposed in cardboard holders using Kodak Industrial AA film, which is a very fine grain film.

RECORDING DENSITOMETER CAPABILITY

Figure 6 shows the positive of a radiograph in which a piece of heavy lead sheeting was placed over a part of the os calcis when the film was taken. The purpose of this figure is to show the range of the recording capability of the Bone Densitometer Assembly. In making the traces of this film, a setting range was established from zero for no light transmission to 9.5 for the maximum light transmission, the latter represented by the area covered by lead.

The aluminum alloy wedge is scanned in its characteristic manner, with the noncorrected raw scan shown in the figure. This wedge trace does not extend over the full scale because the full scale instrument deflection was set up so as to represent the absorption shown by lead.

The bone trace is interrupted when the tracing goes from the bone to the superimposed lead. The difference between the top of the wedge scan and the trace at the position market *lead* on the figure shows the amount of silver which still is on the film and active; and this represents the broad capability of the recording densitometer.

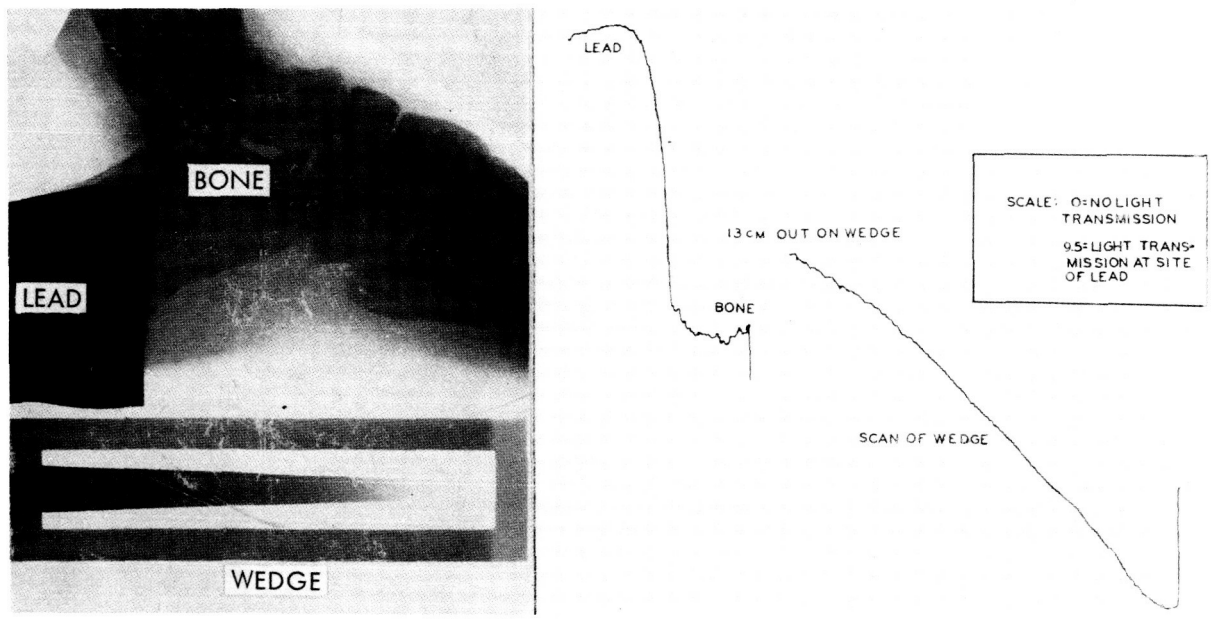

FIGURE 6.—Positive of a radiograph in which a piece of heavy lead impregnated sheeting has been placed over a part of the os calcis of an adult male subject, showing the capability of the recording densitometer in use at the Texas Woman's University.

SELECTION OF RADIOGRAPHIC EXPOSURE CONDITIONS

The X-ray film exposure conditions were selected so that the bone and wedge X-ray images would fall as nearly as possible on the comparatively straight central portion of the Hurter and Driffield characteristic curve for X-ray photographic film.

Influence of Exposure Time

The influence of exposure time on the shape of the noncorrected scan of the 10:1 wedge was studied with kilovoltage held constant at 60 and milliamperage constant at 100, with the results shown in figure 7. It is realized that the curves presented in this figure represent the total effect of the X-ray exposure and the densitometer process, although they give information equivalent to an "H and D" curve and are so called. In this experiment, the effects of four exposure times on the shape of the wedge curve was studied. (The wedge used was the same as that with which an adult os calcis is X-rayed.) The exposure times were 0.4, 0.6, 0.8, and 1.25 seconds. The two traces at the shorter times of exposure showed fairly linear portions in the central part of the uncorrected curve.

As the exposure increased, however, the curve went to the right of the "H and D" curve, with the beginning of the wedge trace becoming progressively flatter. A time of 0.6 second, with the other exposure variables as indicated, was selected for the exposure of an os calcis of an adult male, because the uncorrected curve indicated that the position on the "H and D" curve was favorable and because the time of exposure was more compatible with the thickness of this bone than a shorter time.

For other reasons besides the position of the uncorrected wedge curve on the "H and D" curve, it was deemed important to select a low exposure energy for radiographing the os calcis, which is the most frequently used anatomical site. For example, it is desirable to minimize X-ray changes due to protein. At higher exposure energies a higher proportion of protein is included in the X-ray image than at lower energies. It has been demonstrated in our bed

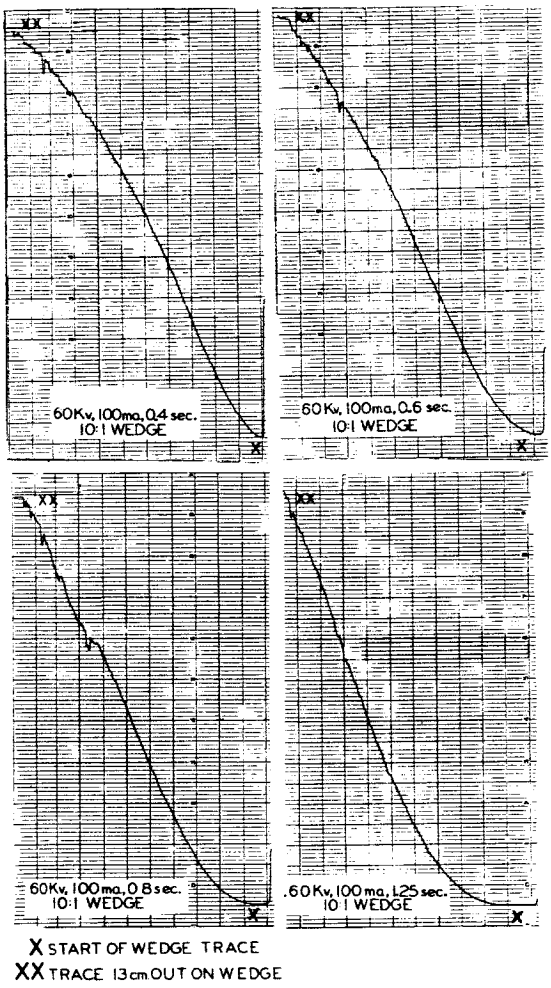

FIGURE 7.—Influence of exposure time on uncorrected os calcis wedge X-rayed at constant levels of kilovoltage and milliamperage.

rest studies that nitrogen is excreted in the urine along with calcium and phosphorus, as seen in figure 8. Some of this could originate from the interstitial protein in bone as well as from the protein of surrounding soft tissue. Karl-Åke Omnell (1957) showed that the mass absorption coefficient of calcium hydroxyapatite at very low exposure energies is 10 times that of protein, while the differential becomes progressively less as exposure energies increase. At very high exposures the mass absorption for this calcium complex becomes almost as low as that for protein, fat, and water.

In order to take advantage of as much of the flattened part of the "H and D" curve as

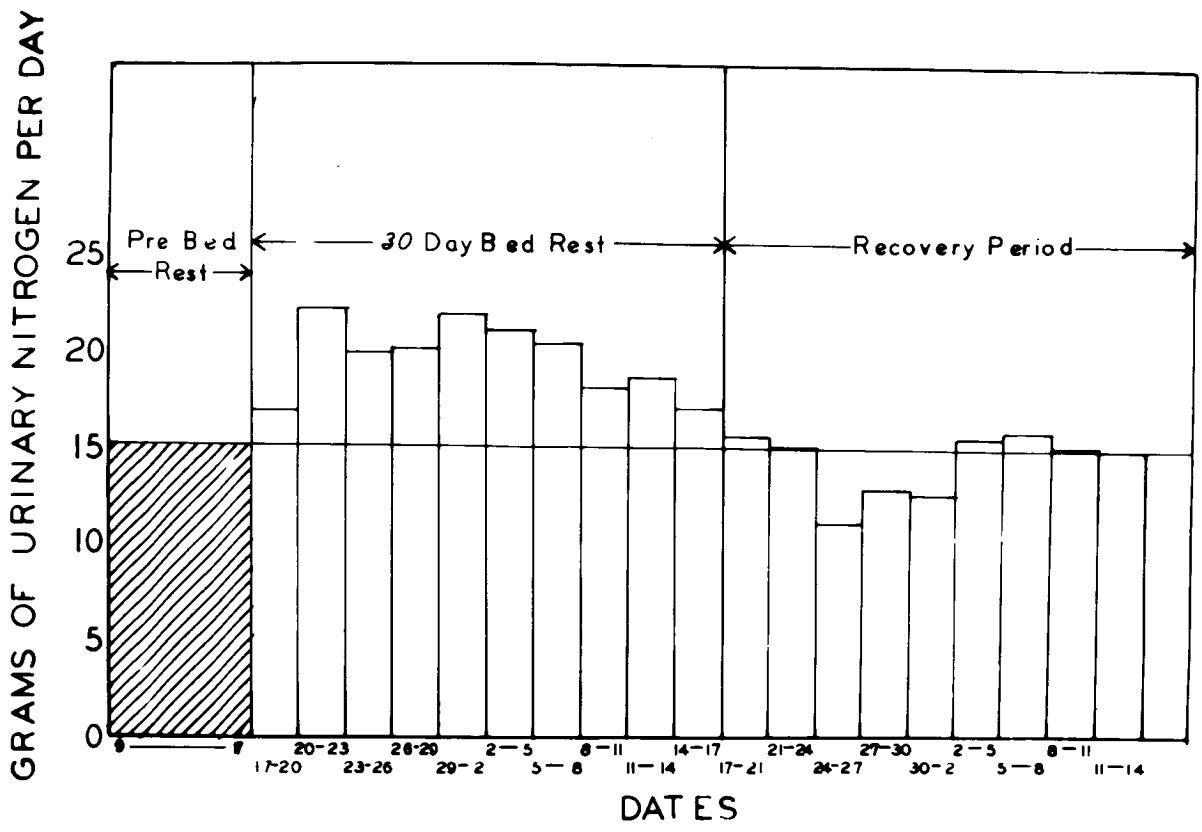

FIGURE 8.—Diagram of daily losses of urinary nitrogen during a 30-day bed rest period, with the preconditioning and reconditioning periods also shown. This illustrates the fact that protein also is lost during bed rest and that as low X-ray exposure energies as possible should be used in order for bone mineral to be predominant in bone mass losses.

possible a somewhat higher exposure energy is needed for the 5 : 1 wedge used for thicker parts of the body.

Figure 9 shows a typical operating range on the Hurter and Driffield curve for a 10 : 1 wedge using an optimum exposure for an os calcis.

A wedge with a 5 : 1 slope, however, which has been found to be more suitable for thicker parts of the body, such as the lumbar spine and the neck of the femur, needs to cover a wider range of exposure than thinner body locations. Somewhat longer exposure times also are necessary for these anatomical sites. The most suitable part of the "H and D" curve for this wedge, therefore, has been found to be the central area between the lower and upper curves. Figure 10 shows a typical operating range for this wedge.

IMPORTANCE OF EXTENT OF COLLIMATION OF X-RAY BEAM IN OBTAINING ACCURACY OF BONE MASS RESULTS

Some of our calibration wedges, particularly those developed for the os calcis and the thicker parts of the human anatomy, are too long to cone the X-ray beam below 12 inches in diameter. The Videx Palmer Radiographic Collimator with which our machine is equipped permits a variety of diameters, with the 12-inch diameter most commonly used, except for the finger and for certain anatomical sites for subhuman primates. For these latter sites, smaller diameters are used.

In order to test the accuracy of the results obtained with a calcium compound X-rayed at

40 inches above the table, with a collimated diameter of 12 inches, a series of experiments have been conducted of which this is an example. The detailed results of the series will be presented when they are completed. Nine

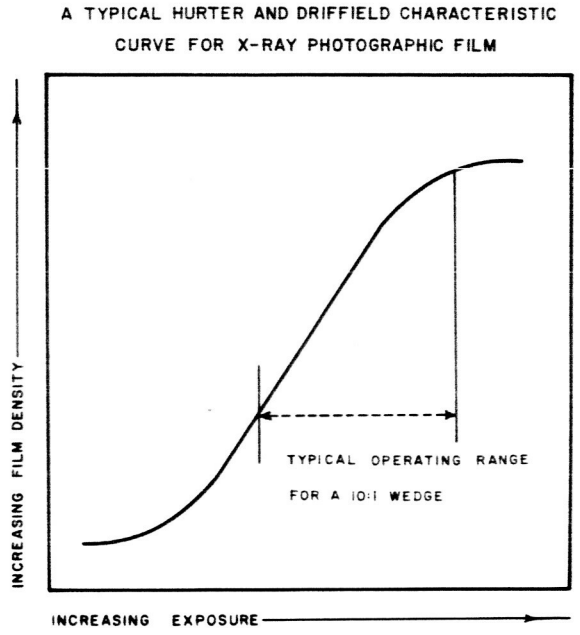

FIGURE 9.—Effective operating range for X-raying a calibration wedge which has a 10 : 1 slope.

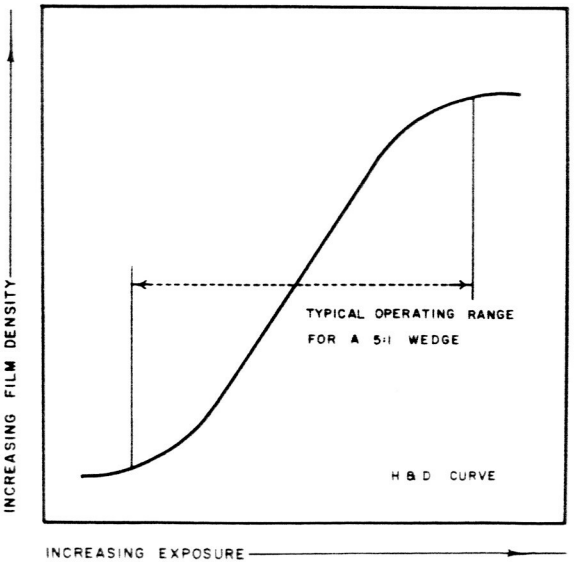

FIGURE 10.—Effective operating range for X-raying a calibration wedge which has a 5 : 1 slope.

FIGURE 11A.—Lateral view of an X-ray positive of an adult human male's left os calcis together with the calibration wedge, within a collimated 12-inch circle which defines the X-ray beam, with the illuminated center spot marked on the os calcis.

small plastic tubes, of 12 mm inside diameter and 4.8 cm deep, were used. One tube remained empty while eight were filled with stepwise quantities of Baker's analyzed calcium hydroxide. Three arrangements of the tubes were X-rayed, as shown in figure 11.

Figure 11 has four parts. Part A consists of a positive of a typical os calcis radiograph with the os calcis calibration wedge in its characteristic location. The illuminated center spot which coincides with the center of the X-ray beam is indicated by a small black spot. The radiograph of the os calcis and the accompanying wedge are used in this figure for purposes of comparing its position within the circle of radiation with the positions of the tubes of calcium hyroxide.

Part B of the figure shows the nine tubes randomly placed, with the tops up, on the one side of the calibration wedge. In Part C, the tubes are arranged in a cluster approximately twice as far from the wedge as was the os calcis in its characteristic location. In Part D, the cluster of tubes has been moved to a distance from the wedge which is approximately twice

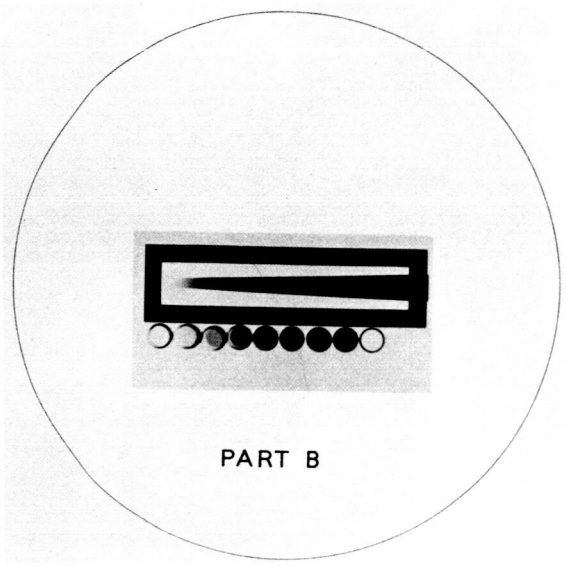

FIGURE 11B.—Xray positive of a series of plastic tubes, one of which was empty while the remainder were filled with stepwise quantities of calcium hydroxide. In this view they were arranged randomly on one side of the os calcis calibration wedge, in the center of the 12-inch collimated X-ray beam. The illuminated center spot of the beam is marked with a black spot.

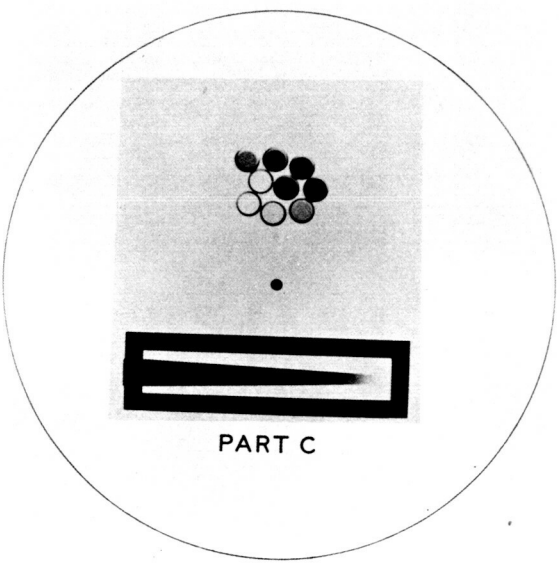

FIGURE 11C.—X-ray positive of the same series of plastic tubes arranged in a cluster inside the same 12-inch circle but situated approximately twice as far from the calibration wedge as was the case with the os calcis in 11A.

as far as the distance between tubes and wedge in Part C. The black spot on Parts B, C, and D again represents the central illumination spot.

In evaluating the quantity of calcium hydroxide in each of the plastic tubes the wedge on each of the three films was scanned on the first recorder of the Densitometer Assembly in the usual manner and then was corrected. Each of the tubes in each of the films was scanned for a distance of 5.0 mm in a portion of the tube where there was no effect from the sides of the containers, with the axis of scan oriented so that the values obtained came only from the calcium hydroxide and the bottom of the containers, with the latter negligible. The results were obtained as integrator counts.

Figure 12 shows the comparative values obtained in terms of integrator counts obtained from the calcium hydroxide in the tubes in Parts B, C, and D, respectively, as the stepwise quantities of calcium hydroxide increased. Part B

FIGURE 11D.—X-ray positive of the same series of plastic tubes arranged in a cluster inside the same 12-inch circle but situated approximately twice as far from the calibration wedge as they were in 11C.

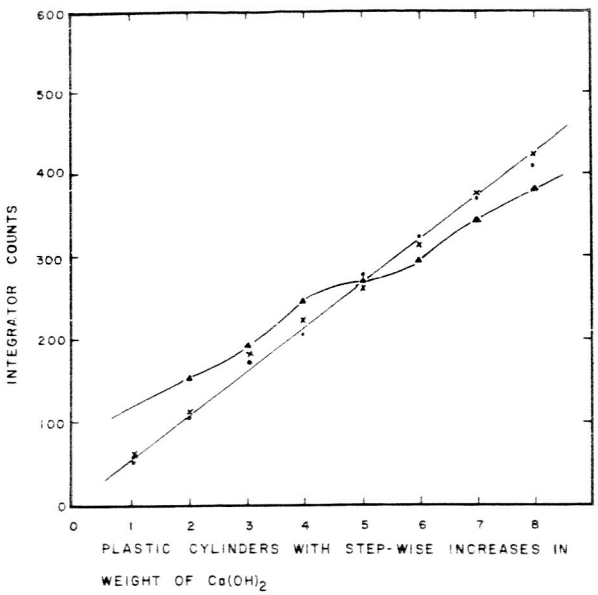

FIGURE 12.—This graph shows the relationship of stepwise quantities of calcium hydroxide in plastic tubes as measured on a quantitative balance and as evaluated by the densitometer in terms of integrator counts. The relationship is shown to be linear for the arrangements in Part B (dots) and in Part C (x's). For Part D this relationship is sporadic, showing that there is a limit to the placement of substances to be evaluated densitometrically if accurate results are to be obtained.

shows that the graph representing integrator counts and quantities of the calcium compound was linear when the tubes were grouped along one side of the wedge. Likewise, the clustered tubes approximately twice as far from the wedge as the os calcis showed linearity with respect to the two factors undergoing comparison.

The group of tubes were situated so far from the wedge that the wedge itself could not have been placed close at hand and still fall within the 12-inch diameter circle.

These findings showed that the placement of an os calcis properly situated with respect to the calibration wedge within a collimated beam 12 inches in diameter could give accurate results. In fact, if it were farther from the wedge than it is customarily placed, accurate results still could be expected. A limit far beyond any locations which we use, however, was found in the case of the third part of the experiment.

792-955 O—66——4

COMPARISON OF EVALUATIONS OF CALCIUM HYDROXIDE BASED ON UNCORRECTED AND CORRECTED TRACES OF THE CALIBRATION WEDGE

Our wedges have been calibrated many times against pure chemicals which are known to be a part of bone mineral. In these calibrations we have used our own technique, correcting the raw, uncorrected wedge curve by the method described earlier in this report as the first step in our densitometric evaluation procedure, and then correcting the raw trace before proceeding.

In order to find whether or not the same results would have been obtained if the raw, uncorrected curve had been used without correction, the following experiment has been performed.

Nine small plastic cylindrical containers of the type used for the above experiment were used in this experiment. They were found to absorb only small amounts of X-ray and not to differ from each other in this respect. They were filled with different stepwise amounts of calcium hydroxide than were used above. The first tube remained empty. The second tube contained 0.129 gram of the compound, with each succeeding tube increased by this amount. Figure 13 shows the radiograph of the nine tubes arranged in random order on the two sides of the calibration wedge. The tubes and wedge were X-rayed at a tube distance of 40 inches, at 60 kV, 100 mA, and 0.4 second.

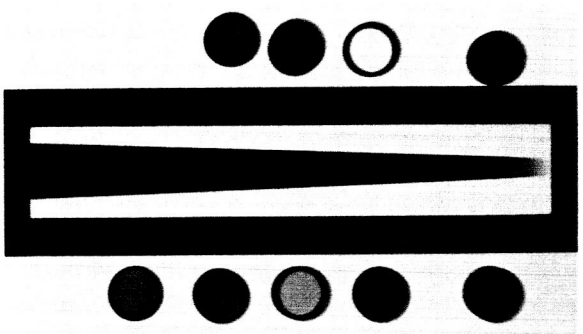

FIGURE 13.—Positive of radiograph of nine plastic tubes of which eight contained stepwise amounts of calcium hydroxide arranged in random order on both sides of the os calcis calibration wedge, with the ninth tube empty.

The densitometric scans were made exactly 5.0 mm in length in the center of the tube image, with the results recorded as integrator counts.

Figure 14 shows two graphs based on weight of the calcium compound plotted against the integrator counts derived from the Densitometer. The graph marked with the x's shows the results obtained when the integrator counts were taken from the procedure in which the initial, noncorrected wedge curve was used. When the wedge curve was corrected according to our procedure with the corrected wedge curve, which is used as a standard for our system, the plot represented by the dots was obtained.

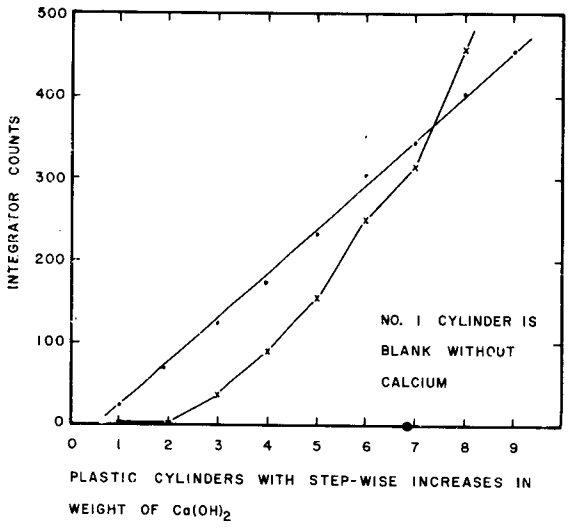

FIGURE 14.—This figure includes two graphs in which the mass of each of a series of calcium hydroxide samples in stepwise quantities as determined on an analytical balance is plotted against integrator counts derived from a densitometric tracing, the integrator being convertible to mass by a simple mathematical formula. The calcium hydroxide samples were in small plastic tubes with one empty.

In the graph delineated by dots, the calibration wedge was traced on the first recorder of the Densitometer Assembly, then was corrected by setting the dials in the potentiometer panel, and then was retraced on the second recorder after which the contents of each individual tube were scanned.

In the second graph, defined by x's, the raw, uncorrected calibration curve was traced, but the correction of this trace was omitted before the calcium hydroxide samples in the tubes were scanned.

The fact that the plot based on densitometer values for the wedge which was corrected is linear with respect to the actual weights of the calcium hydroxide samples, whereas no linearity exists between analytical balance determined mass and densitometer determined integrator counts convertible to mass illustrates the importance of correcting the raw curve of the calibration wedge before making an evaluation of mass densitometrically from X-rays.

The graph based on the corrected wedge curve gave a linear relationship between the integrator counts and the stepwise increase in weights of the calcium compound, while the data derived from the uncorrected wedge curve failed to give stepwise integrator counts for stepwise changes in weights of the calcium hydroxide.

REPRODUCIBILITY OF BONE DENSITOMETER TRACING TECHNIQUE

As a test of the reproducibility of the bone densitometer tracing technique which has been discussed in this report, eight films of Dr. Fred B. Vogt's heel bone were taken within a period of one-half hour. Dr. Vogt monitored the tracing of the films and the analysis of the data which are presented in table I.

The author positioned the subject and supervised the taking of the X-rays, as is done for all X-rays taken in this laboratory. The subject was required to get on and off the X-ray table and be repositioned for each film taken. After being located on the table, a positive of an os calcis film was placed under his foot over the X-ray film, which was encased in a cardboard holder. The purpose of the X-ray positive was to position the foot indentically for each film and to insure that the location of the wedge with reference to the os calcis remained the same each time an exposure was made. The positive was removed before the film was taken. This procedure is followed in all longitudinal studies, with a subject's positive made from the first radiograph taken in a series.

The position of the X-ray tube and of the illuminated center spot which coincides with the center of the X-ray beam was adjusted for each film, with the center light spot made to coincide with the same place on the side of the foot with each repeated position.

The same exposure conditions were used for

TABLE I.—*Integrator Counts Made of Eight Os Calcis Films Processed and Analyzed Three Times for Each Film, with the Eight Films taken of the Same Subject within One-Half Hour, with the Subject Repositioned between Film Exposures*

	Film 1				Film 5		
Segment 1	496	498	506	Segment 1	500	501	502
Segment 2	791	796	782	Segment 2	804	799	800
Segment 3	1037	1041	1041	Segment 3	1053	1058	1054
Segment 4	1240	1237	1253	Segment 4	1264	1269	1239
Segment 5	1287	1281	1302	Segment 5	1311	1314	1310
Segment 6	1279	1275	1294	Segment 6	1306	1304	1306
Segment 7	1232	1239	1240	Segment 7	1241	1245	1240
Segment 8	1230	1226	1229	Segment 8	1232	1238	1243
Segment 9	1265	1264	1254	Segment 9	1265	1278	1269
Segment 10	1223	1231	1225	Segment 10	1229	1236	1249
Total	11080	11088	11126	Total	11205	11242	11212
	Film 2				Film 6		
Segment 1	506	502	497	Segment 1	504	503	505
Segment 2	787	784	787	Segment 2	796	799	798
Segment 3	1034	1039	1033	Segment 3	1048	1051	1051
Segment 4	1241	1236	1234	Segment 4	1259	1253	1268
Segment 5	1280	1294	1274	Segment 5	1301	1290	1310
Segment 6	1286	1301	1281	Segment 6	1300	1286	1297
Segment 7	1222	1229	1227	Segment 7	1247	1245	1245
Segment 8	1234	1239	1235	Segment 8	1243	1240	1243
Segment 9	1271	1269	1251	Segment 9	1270	1266	1278
Segment 10	1239	1240	1255	Segment 10	1243	1241	1247
Total	11100	11133	11074	Total	11211	11174	11242
	Film 3				Film 7		
Segment 1	501	498	497	Segment 1	503	503	502
Segment 2	783	781	800	Segment 2	803	810	806
Segment 3	1046	1053	1041	Segment 3	1041	1046	1044
Segment 4	1254	1247	1237	Segment 4	1241	1244	1243
Segment 5	1299	1283	1291	Segment 5	1294	1310	1302
Segment 6	1286	1277	1290	Segment 6	1291	1300	1297
Segment 7	1240	1242	1231	Segment 7	1231	1236	1239
Segment 8	1237	1234	1236	Segment 8	1229	1233	1231
Segment 9	1275	1265	1258	Segment 9	1274	1265	1268
Segment 10	1228	1236	1234	Segment 10	1244	1241	1234
Total	11149	11116	11115	Total	11151	11188	11166
	Film 4				Film 8		
Segment 1	507	502	502	Segment 1	496	503	507
Segment 2	800	801	804	Segment 2	797	807	791
Segment 3	1041	1041	1039	Segment 3	1038	1029	1058
Segment 4	1260	1262	1237	Segment 4	1236	1240	1265
Segment 5	1304	1291	1296	Segment 5	1300	1304	1314
Segment 6	1295	1287	1301	Segment 6	1294	1311	1302
Segment 7	1240	1243	1230	Segment 7	1230	1232	1239
Segment 8	1249	1240	1236	Segment 8	1240	1238	1243
Segment 9	1263	1260	1278	Segment 9	1267	1275	1275
Segment 10	1237	1228	1250	Segment 10	1228	1239	1252
Total	11196	11155	11173	Total	11126	11178	11246

each film—namely, 60 kV, 100 mA, and 0.6 second. All films were processed in the developer at the same time. The subject was protected during all exposures by lead shielding placed over his body, covering all areas except the foot, with the shielding replaced for each new positioning.

The results in the table represent the analysis of the central section of the os calcis (see fig. 3). The section was segmented as it was traced by interrupting the scan and taking an integrator count for 10 separate segments in each bone scan, as shown in the cross-sectional view through which this scan is traced (see fig. 5).

On each film the os calcis was analyzed three separate times, with the film completely repositioned in the densitometer before each analysis, and with the wedge scanned, the uncorrected wedge trace corrected, and the proper section of the bone scanned independently for each trial. The positions of the scan on os calcis and on the wedge were located on each film by needle pricks at the posterior-anterior extremities of the bone scan position, but outside of the bone. The wedge extremities were also located in the same manner. Exact locations of the landmarks for the scans were found for the various films by superimposing the films one at a time on top of the initial film and locating the positions of the needle pricks from this first film.

Detailed statistical analysis of these data has been prepared and is being presented elsewhere for publication. To give some evaluation of the range of error that can be expected for the total process of taking the film, developing the film, and analyzing the film on the densitometer, an analysis of variance was performed on the sums of the counts for the segments for each of the three analyses made on the eight films. The data are presented in integrator counts.

The 99% confidence level is expressed in mean densitometer counts of 11,160, with a range of counts above and below this mean approximating 0.75% of the total counts. Thus it could be said that the "error" which is found between the films as analyzed is represented at the 99% confidence level by a span or range of error of 0.75% above and below the mean for a total range of 1.5%.

ACKNOWLEDGMENTS

The author is deeply indebted to those who, through the years, have cooperated in the development of equipment and the standardization of method for the measurement of bone mass by the densitometric scanning of X-rays. Especial gratitude is due to Ernest Axman, Hughes Daniel Trapp, W. Lewis Shelter, and the late Walter N. Brown, Jr., for assistance in the development of the earlier models of our scanning densitometers, and to George P. Vose and the late James Donald Nelson and his colleagues at the Leeds and Northrup Company for collaboration in the design and building of the Bone Densitometer Assembly which we are now using.

Dr. Fred B. Vogt, a staff member of the Texas Institute for Rehabilitation and Research of the Baylor University College of Medicine (Houston), has been most helpful during the past 2½ years by serving as a consultant to our group at the Texas Woman's University in our NASA bed rest studies and, particularly, by cooperating in experiments designed to test the validity of our radiographic bone densitometric procedures.

REFERENCES

CANTEROW, ABRAHAM; and SHEPARTZ, BERNARD: Biochemistry, Third ed., W. B. Saunders Company, Philadelphia, 1962.

MACK, PAULINE BEERY; VOSE, GEORGE P.; and NELSON, JAMES DONALD: New Developments in Equipment for the Roentgenographic Measurement of Bone Density, Am. J. Roentgenol., Radium Therapy, Nucl. Med., vol. 82, 1959, p. 303.

NELSON, JAMES DONALD; MACK, PAULINE BEERY; and VOSE, GEORGE P.: New Design of a Linearizing Recording Densitometer. Rev. Sc. Instr., vol. 20, 1958, p. 316.

OMNELL, KARL-ÅKE: Quantitative Roentgenologic Studies on Changes in Mineral Content of Bone in Vivo. Acta Radiol., Suppl. 148, 1957.

Factors Affecting the Precision of Radiographic Densitometry of the Lumbar Spine and Femoral Neck

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The increasing interest in the measurement of bone mineral content by X-ray or gamma-ray absorptiometry is clearly indicated by the numerous investigations currently underway involving applications of the various techniques. Since the science is not a new one, it is perhaps surprising that it has not yet been developed to highest perfection. Certainly far more progress has been made in more formidable technological tasks. Television, for example, was developed to a reasonably good state in less than three decades of experimentation, and manned space vehicles were orbiting the earth within a single decade of organized research. On the other hand, although X-ray utilization in the medical sciences is an old art (scientists were experimenting with quantitative evaluations of skeletal density as early as 1897), we are still somewhat uncertain of its ultimate capacity or even its present state of development.

Since the earliest attempts with bone densitometry, several methods have been developed using both polychromatic and monochromatic beams with X-ray film detectors. Recently, monochromatic photon absorptiometry with scintillation detection has been applied in estimating the ash content of certain bones of the appendicular skeleton. Admittedly, the substitution of collimated monochromatic beams in X-ray and gamma-ray bone absorptiometry reduces both the scattering error and the range of effective mass absorption coefficients of the component elements. A chief disadvantage of the monochromatic beam system, however, is the accompanying decrease in intensity which requires relatively long exposures even for such relatively thin body parts as the wrist and hand.

Although appendicular bone densitometry is a useful investigative tool, its major disadvantage is that it may not be a sensitive reflector of changes in the axial skeleton, which includes areas most significantly involved in osteoporosis and other metabolic skeletal disorders. This disadvantage, and the consequent inadequate application of appendicular densitometry, probably accounts for the somewhat disappointing contribution thus far of bone densitometry to our knowledge of bone metabolism. Although X-ray densitometry of bones of the extremities is capable of good reproducibility using current methods, it may not be possible for any X-ray of gamma-ray absorption technique to detect the minute calcium accretion or resorption occurring during periods of one or two weeks within a single bone of the appendicular skeleton. It is well known, however, that bone demineralization can be detected visually at earlier stages in the axial skeleton than in the appendicular skeleton, and apart from the technical disadvantages, the spine has long been considered the densitometric area of choice. The major technical problem that has hindered vertebral densitometry is the complexities resulting from the effects of the superimposed, large, and variable amounts of surrounding soft tissue and

sometimes the nonuniform bowel contents on the bone image. However, it is the cylindrical vertebral column that is usually evaluated when skeletal density is clinically assessed because it is frequently in the spine where a decreased mineral content first occurs, where the decrease in mineral content is ultimately the greatest, and where the consequences of decreased bone tissue are clinically more significant.

This report provides the results of some intensive experimentation on the effects of numerous radiographic variables on densitometry of the lumber spine and femoral neck. Possibly the causes of inconsistencies among the earlier studies by various investigators using other anatomical sites can be traced to some of the effects described in this report. We are well aware that some of our first attempts in vertebral densitometry were handicapped by a lack of knowledge of just how significant some of the variants were. At the Lahey Foundation in Boston, for example, an application of the newest techniques is expected to clear up some of the inconsistencies which were noted during the earlier clinical applications.

The limits dictated by the numerous controlling factors are now clearly obvious. Hence, the overall precision of radiographic densitometry should be increased if the limitations imposed by the factors studied in this investigation are adhered to rigidly.

GENERALIZED TECHNIQUE FOR HUMAN VERTEBRAL AND FEMORAL DENSITOMETRY

Although the methods applied in radiographic densitometry of the lumbar spine and femoral neck are essentially the same as those utilized in densitometry of the appendicular skeleton, the increased masses of obscuring tissue requires the rigid application of highly standardized techniques.

In a strictly abbreviated presentation, these include the use of collimated beams, shielding the X-ray table, subject, and calibration standard with sheet lead and lead rubber to prevent "undercut" scattering, and precise application of exposure and darkroom processing methodology. The practice of exposing the body part and calibration wedge side-by-side (as in appendicular bone densitometry) has been modified. In the case of the femoral neck, the wedge is placed on a scribed plastic shield overlying the hip in such a way that the X-ray image of the wedge is projected through the soft tissue of the thigh immediately adjacent to the bone.

In densitometry of the lumbar spine, the beam is first centered over vertebra L3, which is projected onto the top one-half of a 14 x 17-inch film, and immediately thereafter centered upon the aluminum-plexiglass phantom, which is projected onto the bottom one-half of the film. In all cases, replicate exposures are made in order to detect possible energy variations between exposures of bone and calibrating material.

The method requires the use of high speed films and calibrated intensifying screens, reciprocating bucky diaphrams, and rigid consideration of all factors discussed in this report.

THEORETICAL

Part I. General

The theoretical aspects of absorptiometry of homogeneous materials are well known, so will be dealt with only briefly in this report. If the absorber consists of a single element (or group of elements in constant proportionality to provide a mean mass absorption coefficient) the mass of material is proportional to its absorbance of an X-ray beam of known quality and intensity. In the case of an absorber having two components of widely different absorption coefficients, the ratio of the two components can be determined from the X-ray attenuation, providing either the total mass or volume of the absorber is known. Bone falls into this category, since it is composed of two fractions—mineral and water organic matrix—each having widely separated absorption coefficients.

At an effective wavelength of 0.413 Å, for example, the mass absorption coefficient of the organic fraction of bone is approximately 1.30, while that of the inorganic fraction is 14.8. At this wavelength, therefore, the organic-inorganic absorption ratio

$(\mu/\rho)_o/(\mu/\rho)_i$ is 0.09. At a higher energy wavelength of 0.124 Å the ratio is 0.36. These data indicate that the mean absorption coefficient of bone will vary in accordance with the wavelength of X-radiation and the relative proportions of organic and inorganic fractions in the absorbing mass. With the wavelength remaining constant, the organic-inorganic ratio of the material then establishes its mean mass absorption coefficient.

To validate such an assumption, it must be shown that variations in the ratios of the component elements of the organic and inorganic fractions do not alter the absorption by each fraction as a whole. In this regard, the inorganic fraction of bone will first be considered.

Although the chemical structure of bone mineral is not completely understood, calcium and phosphorus obviously comprise its major absorbing elements, with the calcium to phosphorus ratio in bone generally accepted to vary from 1.5 to 2.0. To determine the change in transmittance accountable to varying ratios of calcium and phosphorus in the absorber, mixtures of calcium carbonate and $CaH_4(PO_4)_2 \cdot H_2O$ were prepared in such a way that the calcium-phosphorus ratio ranged from 0.64 to 3.16. Within these limits the transmittance of the mixture (at 40 kV) was changed by less than 1%. Although any change in the Ca : P ratio would be expected to have a more pronounced effect than this on total absorption, the absorption accountable to the oxygen component of bone mineral tends to subordinate the effects of such ratio changes since oxygen accounts for approximately 50% of the weight of the molecule.

Variations in the relative proportions of the organic components have an even smaller effect upon the mean absorption coefficient. The organic fraction in dry bone contains between 90 and 96% collagen or collagen-like proteins, and a few percent of fat. In the case of a low energy X-ray beam, the absorption coefficient of fat is approximately 70% that of protein. Because the absorbing properties of protein and fat are so closely related, the expected variations in the relative quantities of these two components would be of no significance.

Part II. Soft Tissue Corrections

At the present time a few investigators are using two widely different wavelengths to separate the soft components from mineral in X-ray densitometry. This approach, although theoretically sound, does not invalidate the use of either a single wavelength or a polychromatic beam. As shown in the previous section, the mass absorption coefficient of bone will vary in accordance with the effective wavelength of the X-radiation and the relative proportions of organic and inorganic fractions. With the wavelength remaining constant, the inorganic/water-organic ratio then establishes its mean mass absorption coefficient. For example, at an energy of 45 keV the mass absorption coefficients of hydroxyapatite, protein, water, and fat are 0.74, 0.23, 0.24, and 0.22 respectively. Since the fat component of bone is fairly low, the average mass absorption coefficient of the bone is quite close to 0.23—which is significantly lower than the 0.74 of the mineral fraction. What we are measuring with either a single wavelength or a polychromatic beam, therefore, is the weight concentration of bone mineral to its water-organic matrix.

The foregoing statements are intended to defend the validity of separating bone mineral from its soft fractions. In practice, however, the soft tissue overlying the bone is actually of greater significance than the soft tissue within the bone.

Various attempts have been made to correct for soft tissue absorption as a source of error in quantitative X-ray measurements. The current method used by the writer is essentially the exposure of a calibration wedge through the same thickness of tissue (or tissue-simulating plastic) as that overlying the bone itself. In the case of the femoral neck, the wedge is placed over the subject's hip in such a way that its image is projected beside the bone with approximately the same thickness of soft tissue complicating each image. In the case of the third lumbar vertebra the calibration wedge is exposed through a thickness of plexiglass plates equivalent to the lateral thickness of the patient. Admittedly, plexiglass does not have precisely the same X-ray absorption as all soft tissues,

but the difference in each radiograph can be determined by densitometric comparison of plexiglass and the soft tissue adjacent to the bone, and the scan profile is either elevated or depressed accordingly. It is emphasized that on followup films of the same subject it is necessary to use the same number of plexiglass plates that were used initially.

In the case of the femoral neck, since individual musculature varies within short distances, a similar wedge scan adjustment must be made on each densitometric scan of the bone.

THE ABSORPTION STANDARD

In order to investigate reproducibility as affected by radiographic factors alone (uncomplicated by anatomical effects) it was first necessary to construct a standard absorber from components having similar X-ray scattering and absorption characteristics as bone and soft tissue, and which could be positioned under the X-ray beam in a reproducible manner.

The standard absorber simulating bone and tissue used in these tests is illustrated in figure 1 and is essentially a machined 5:1-slope aluminum alloy wedge embedded in a plexiglass plate of 5.8-cm thickness. The composition of the alloy is 93.4% aluminum, 4.5% copper, 1.5% magnesium, and 0.6% manganese. Embedded also in the plate and immediately adjacent to the wedge is a simulated "bone" composed of 40% human bone ash (575° C) and 60% casein to simulate the soft tissue component. This material is compressed into a 1.3-cm hole drilled through the plate. In use, the absorber is placed in the X-ray field with the central beam projected at a point midway between the wedge and simulated bone. The total thickness of the absorber can be varied by stacking additional plexiglass plates of 1.2-cm thickness each above and below the central plate containing the calibration wedge and bone ash. The entire assembly is placed upon a 15 x 18-inch lead sheet (⅛-inch thickness), having a rectangular section removed with dimensions somewhat smaller than those of the plates. A rectangular lead sleeve is slipped down over the stacked plexiglass plates to reduce undercut scattering as much as possible.

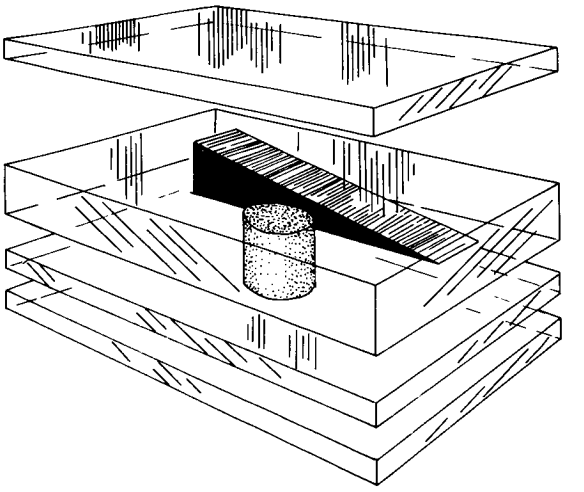

FIGURE 1.—Standard absorber used in the described tests consisting of an aluminum alloy wedge and bone ash embedded in Plexiglas.

The total thickness of plexiglass used in all exposures made during these investigations was 17.8 cm, with five plates being used above and below the central plate.

Figure 2 shows the mass absorption coefficients of bone ash (considered to be hydroxyapatite, $Ca_{10}(PO_4)_6 \cdot (OH)_2$), the aluminum alloy standard, and the three soft components of bone. It can be seen that the aluminum alloy used has absorption coefficients somewhat lower than those of hydroxyapatite throughout the energy range shown. These absorption coefficients could be brought closer together by use of an alloy containing slightly more of one of the heavier elements. This slight difference in X-ray absorption characteristics between bone mineral and the alloy, however, probably does not cause serious errors since the same kilovoltage is used on followup films of the same individuals.

KILOVOLTAGE EFFECTS

Part I. General

Since the ratios of X-ray absorption coefficients between bone and soft tissue and between bone and aluminum alloy change in accordance with changes in the effective wavelength of the beam, it is obvious that the kilovoltage must be controlled rigidly in radiographic bone densitometry. In addition, the kilovoltage effect

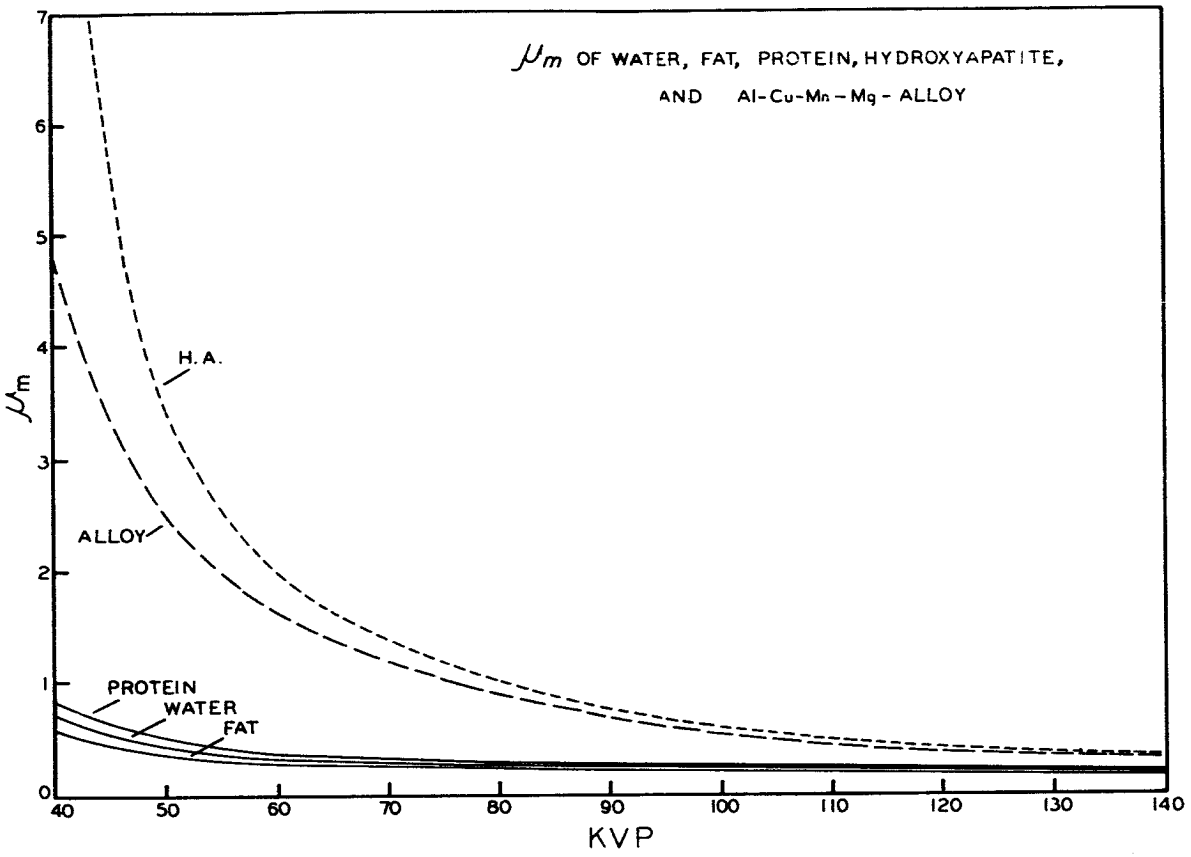

FIGURE 2.—Mass absorption coefficients of hydroxyapatite, aluminum alloy, and three soft tissue components.

appears to be more pronounced when fluorescent screens are used than when nonscreen films are exposed directly to the beam in cardboard holders. Figure 3 illustrates that the X-ray aluminum equivalency decreases as kv is increased (with the MAS adjusted to maintain constant film background density). Within the 10-kV range from 56 to 66 the apparent aluminum equivalency values ranged from 0.83 to 1.00—a change of approximately 21%, or a 2% change per kilovolt. Within a narrower range of 58–62 kV the total error was 4.2%, or about 1% per kilovolt. Since it is known that kV variations of 10% or more exist among different X-ray units, it is possible that aluminum equivalency errors of 30% may occur when one X-ray unit is substituted for another without cross-calibration compensation. The following part of this section of the report describes a method of cross-calibration which has made it possible to reduce an initial error of 31% on an uncalibrated unit to 1.7% after calibration.

At higher beam intensities the error accountable to kV variations is decreased. This effect is illustrated by figure 4 showing a 10.2%/kV error at low MAS (80–125), a 5.2%/kV verror at intermediate MAS (150–250), a 3.0%/kV error at high MAS (250–350), and a 1.4%/kV error at the highest MAS of 350–500.

Although the reproductibility is increased at high MAS, it is possible that the overall accuracy is somewhat diminished in accordance with decreased sensitivity in densitometry of the darkest radiographs. In radiographic densitometry of the adult lumbar spine in lateral projection, the correct kilovoltage is best determined by multiplying the lateral body thickness (cm) at the level of vertebra L3 by a factor of 3. The same factor is also used with the AP projection of the femoral neck as measured in supine position.

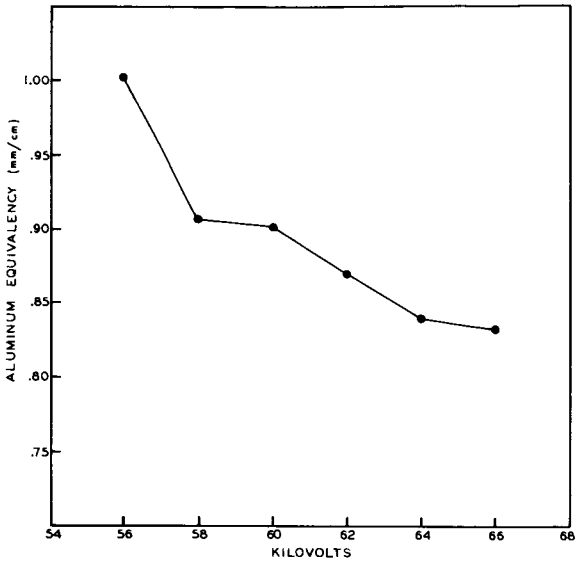

FIGURE 3.—Relationship of apparent X-ray aluminum equivalency with kilovoltage.

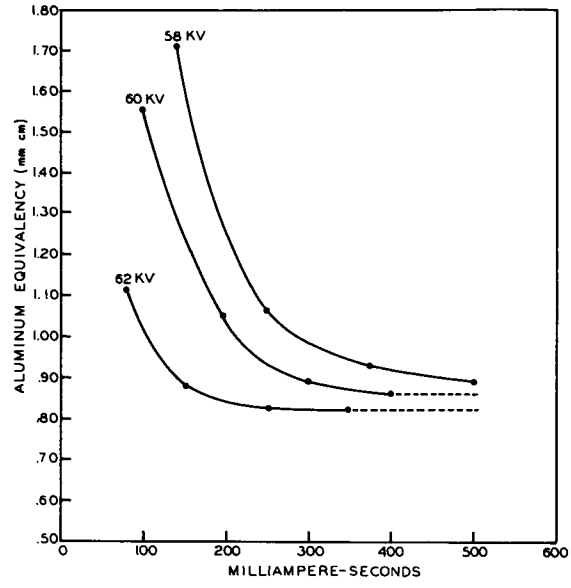

FIGURE 4.—Relationship of apparent X-ray aluminum equivalency with intensity of exposure at 58, 60, and 62 kV.

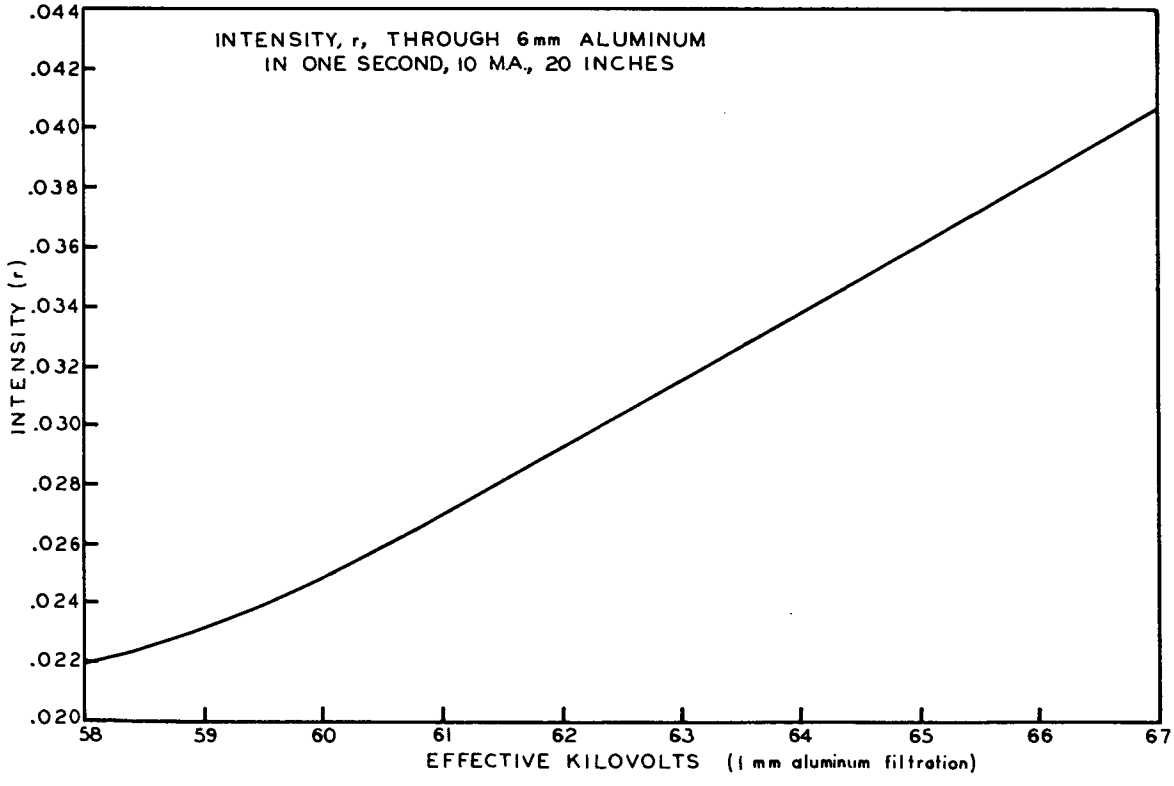

FIGURE 5.—Relationship of radiant intensity with kilovoltage at specific instrument settings on T. W. U. General Electric X-ray unit.

Part II. Cross Calibration of Separate X-ray Units

It has been possible to calibrate the effective kv of other X-ray units with the central unit at Texas Woman's University by means of a calibration curve relating kV with the transmittance through a standard aluminum filter under a specific X-ray intensity. Figure 5 indicates that the energy-intensity variants of the central T.W.U. unit are related by a curvilinear function. For example, on the central unit the X-ray transmittance at 64 kV through a standard 6-mm aluminum filter at 10 MAS and 20 inches distance is 0.034 r. In comparing an uncalibrated unit against the T.W.U. unit to obtain a corresponding 64 kV, a 10-MAS exposure is made on a 0.02 r Victoreen ionization chamber resting in a lead boat which absorbs backscattered rays. In accordance with the calibration curve, a 0.040–r exposure would indicate an actual X-ray energy of 67 kV. In order to align the latter unit with the central unit, a series of exposures must be made until a transmittance of 0.034 r is obtained under the specified conditions. The result indicated kV would then be used as the equivalent of 64 kV on the central unit.

Preliminary radiographs of the plexiglass-aluminum-bone ash phantom made on five X-ray units in three locations, without any attempt for cross-calibrating, yielded widely different values as shown in ascending order in table I. The maximum difference of 31% (units 1 and 5) proves that it would be fallacious to attempt to compare results obtained with two uncalibrated units.

TABLE I.—*Aluminum Equivalencies Obtained on Five Noncalibrated X-ray Units at Same Indicated Technique (60 kV, 100 MAS, 36 inches, 3-mm aluminum filtration, Cronex I film in Patterson high-speed screens)*

Unit No.	Model	Site	Aluminum equivalency
1	Picker 200 ma	Flow Hospital	0.718
2	G.E. 300 ma	Flow Hospital	.746
3	Picker Aeromax	Finlay-Lockwood	.925
4	G.E. KX8	T.W.U.	.980
5	G.E. 11D54	Finlay-Lockwood	1.010

The instrument which provided the lowest value (unit No. 1 at Flow Hospital) was selected for calibration with the T.W.U. unit. Exposures were first made at 58 and 60 kV on the T.W.U. unit and then the standard absorber was reassembled on the unit at Flow Hospital. Previous calibration exposures, using the described ionization chamber method, had shown that dial settings of 54 and 56 kV on the Flow Hospital unit corresponded with indicated kilovoltages of 58 and 60 on the T.W.U. X-ray unit. The comparative results obtained after cross-calibration of the two units are shown in table II. All technique factors except kilovoltage remained constant in this experiment. It is clearly indicated that, with careful prior calibration using the described ionization chamber method, quite accurate kilovoltage compensations can be applied for comparative purposes when different X-ray units must be used.

TABLE II.—*Comparative Aluminum Equivalence Values Derived from Two Separate X-ray Units after Kilovoltage Compensation*

Test No.	Flow Hospital Unit	T.W.U. Unit	Difference between values
1	0.990 (54 kV)	0.978 (58 kV)	1.2%
2	0.939 (56 kV)	0.954 (60 kV)	1.7%

INTENSITY OF EXPOSURE

With effective X-ray wavelength, emulsion characteristics, and processing factors remaining constant, the "blackness" of the X-ray image is controlled by the intensity of the X-ray beam. The intensity, in turn, is the result of combined milliamperage, exposure time, and focal-film distance. It has been a generally accepted principle in diagnostic radiography that intensity changes accountable to changes in tube-film distance can be compensated for by varying the milliamperage or exposure time without affecting the resulting intensity—supposedly by reducing or increasing the product of ma and time in accordance with the square of the change in tube-film distance. We have found that this formula is not acceptable within the rigorous requirements of bone densitometry. With the tube-film distance, kilovoltage, and filtration remaining constant, however, it has been found that the MAS may be changed within fairly large limits without causing substantial changes in X-ray aluminum equivalency. Figure 6 indicates that the apparent aluminum equivalency of the standard Plexiglas-aluminum-bone ash phantom increases with increasing MAS. With the phantom used in these tests, having the same thickness of the human hip (17.8 cm), the optimum exposure was determined to be 275 MAS at 60 kV and 48 inches using Dupont Cronex I film and Patterson high-speed screens. From this optimum exposure it was found that MAS could be increased or decreased by 30% without causing errors in reproducibility greater than 1.8%. Further increases in MAS beyond 30%

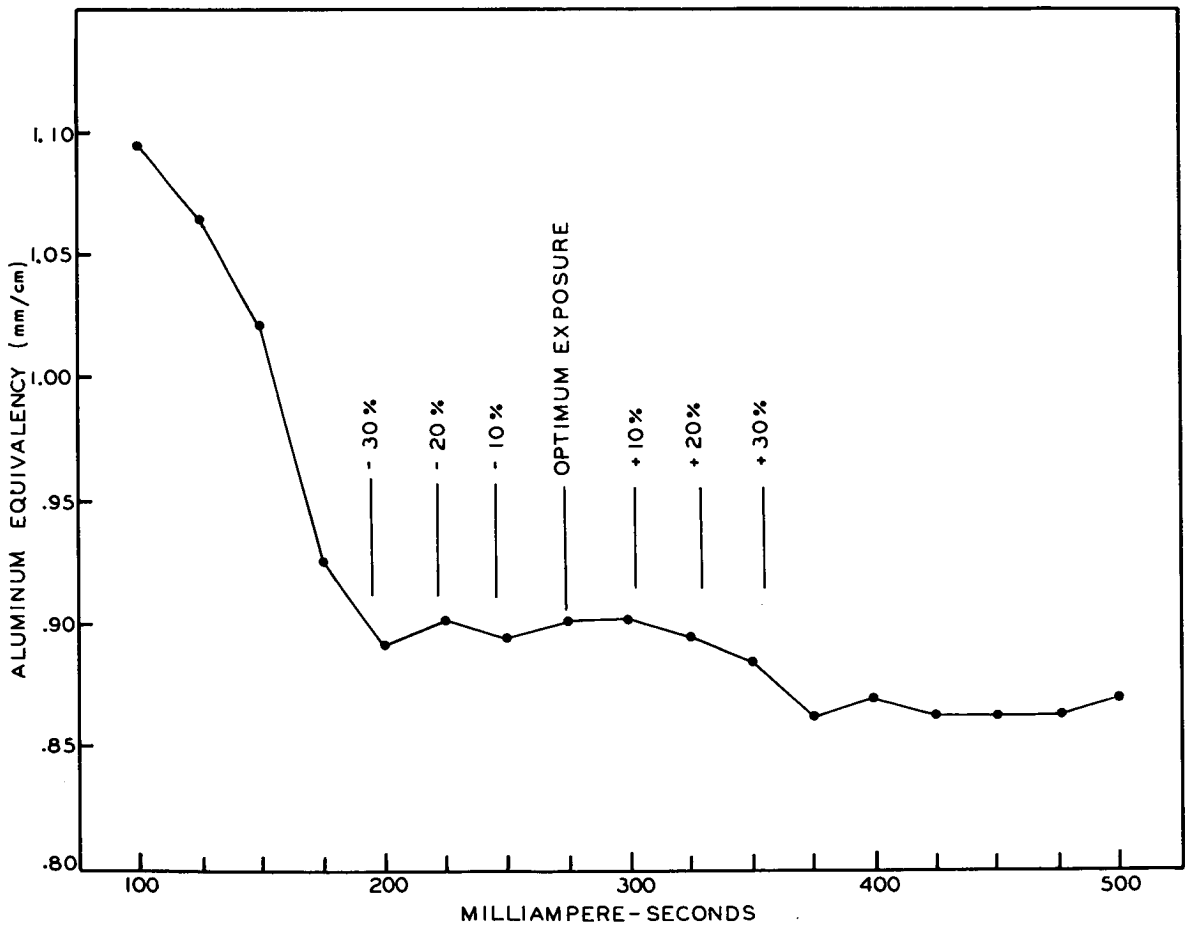

FIGURE 6.—Relationship of apparent aluminum equivalency with intensity of exposure.

from the optimum resulted in some reduction of the aluminum equivalency value (4.2% with a 60% MAS increase). On the other hand, a decrease in MAS below 30% resulted in severe inaccuracies. The graph also indicates the presence of a plateau as the MAS is increased with sufficient length so that minor changes in MAS will not significantly affect the X-ray aluminum equivalency.

BEAM FILTRATION

Although the effective wavelength of the beam is shortened as the aluminum filtration is increased, this variable is easily controlled, so it has not been studied extensively in this investigation.

Figure 7 indicates that the aluminum equivalency value is apparently increased in accordance with increasing filtration by about 3.5% per mm of aluminum up to 3 mm, followed by a more pronounced increase at 4 mm. Since the wavelength of the beam is effectively shortened as the soft waves are removed by filtration, it would be expected that the apparent aluminum equivalency would be decreased with added filtration as indicated by figure 3. Since this does not occur, however, it is obvious that the accompanying overall intensity decrease is the predominating factor which causes the apparent density increase with the heaviest filtration. The aluminum filters furnished by the manufacturers of X-ray equipment are ordinarily machined to close tolerance, thus it is doubtful that variations in aluminum filtration are a significant factor in X-ray densitometry as long as the same thickness is used repeatedly. The window of the X-ray tube itself has an inherent filtration, but this should be insignificant at the relatively high energies used in the technique.

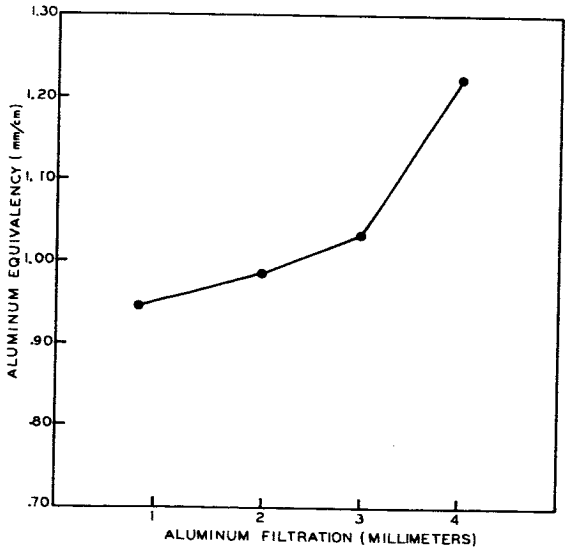

FIGURE 7.—Effect of increasing aluminum filtration on apparent aluminum equivalency.

FILM PROCESSING FACTORS

During the course of these investigations on reproducibility of X-ray aluminum equivalency on living subjects it became apparent that although duplicate films of the same subject exhibited good reproducibility when exposed and processed on the same day, a pair of films exposed a day or so later customarily showed good agreement with each other but often showed poor agreement with the previous films. The observed differences were occasionally as high as 15%.

An experiment was designed to determine the effects of day of development upon the apparent bone density. First it was necessary to determine if films exposed within a 5-minute interval would, after being separated into two groups, yield the same film-to-film variations when developed simultaneously or when developed individually at 24-hour intervals. For this experiment ten separate exposures were made of the aluminum-plexiglass-bone ash phantom within a 5-minute interval. Six films were developed immediately, and four were retained for development at consecutive 24-hour intervals. Figure 8 indicates the apparent aluminum equivalency of each of the ten films, with 1–6 being developed simultaneously while films 7–10 were processed at the same time of day for four consecutive days. The maximum difference among the films developed simultaneously was 2.7%, with a 1.3% deviation from the mean value. However, in the case of the films exposed at the same time but retained for day-by-day development, the maximum variation was 15.4%, with an average deviation from the mean of 3.8%.

Films 9 and 10 on the graph were exposed together but developed one day apart with an in-

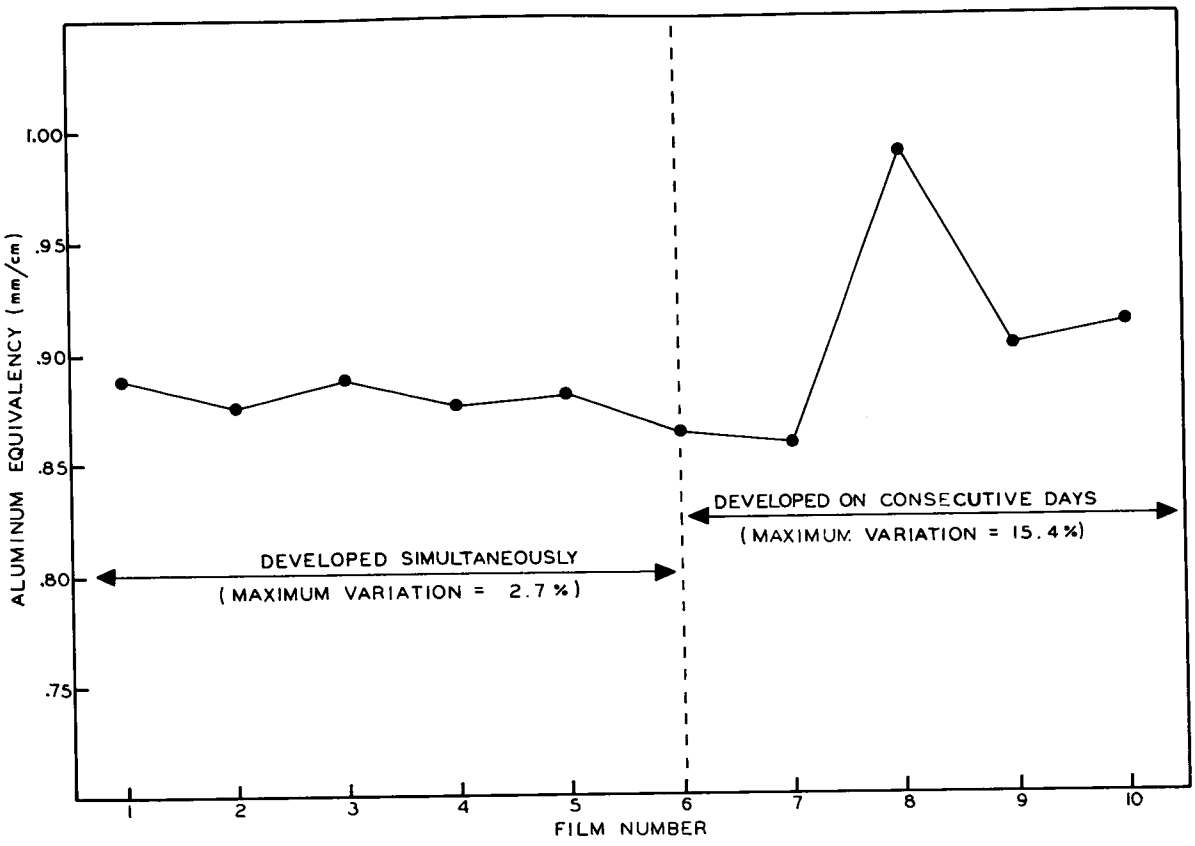

FIGURE 8.—Variations in apparent aluminum equivalency among films exposed together and developed simultaneously, and films exposed together and developed at consecutive 24-hour intervals.

termediate change of X-ray processing solutions. The difference between their respective aluminum equivalencies of 0.901 and 0.913 is 1.3%, indicating that age of X-ray processing solutions was not a significant factor in this particular experiment.

Although the experiment does not prove conclusively that changes in temperature or composition of the X-ray processing solutions caused day-to-day fluctuations in sensitivity, it does indicate that the chances of consistency would be improved by developing all films at one time, preferably in developer prepared fresh daily.

INSTRUMENT AND PROCESS REPRODUCIBILITY

The photometric instrument used in these evaluations consists essentially of a Knorr-Albers microphotometer (manufactured by the Leeds and Northrup Company) and a Speedomax G pen and ink recorder. Both instruments have been modified for the specific application of bone densitometry. The instrumentation and computation processes have been found to be highly reproducible in multiple evaluations of the same radiographs. However, as in all radiation measuring devices with which the writer is familar, an occasional spurious error will occur. For this reason each film is routinely evaluated twice and if the difference in values is within 2% for the femoral neck, or 3% for the third lumbar vertebra, the average of the two values is reported as aluminum equivalency.

Figure 9 illustrates the frequency distribution of errors in replicate evaluations of radiographs of the 84 femoral necks and 124 third lumbar vertebrae produced in this study. A comparison of the slopes indicates that the

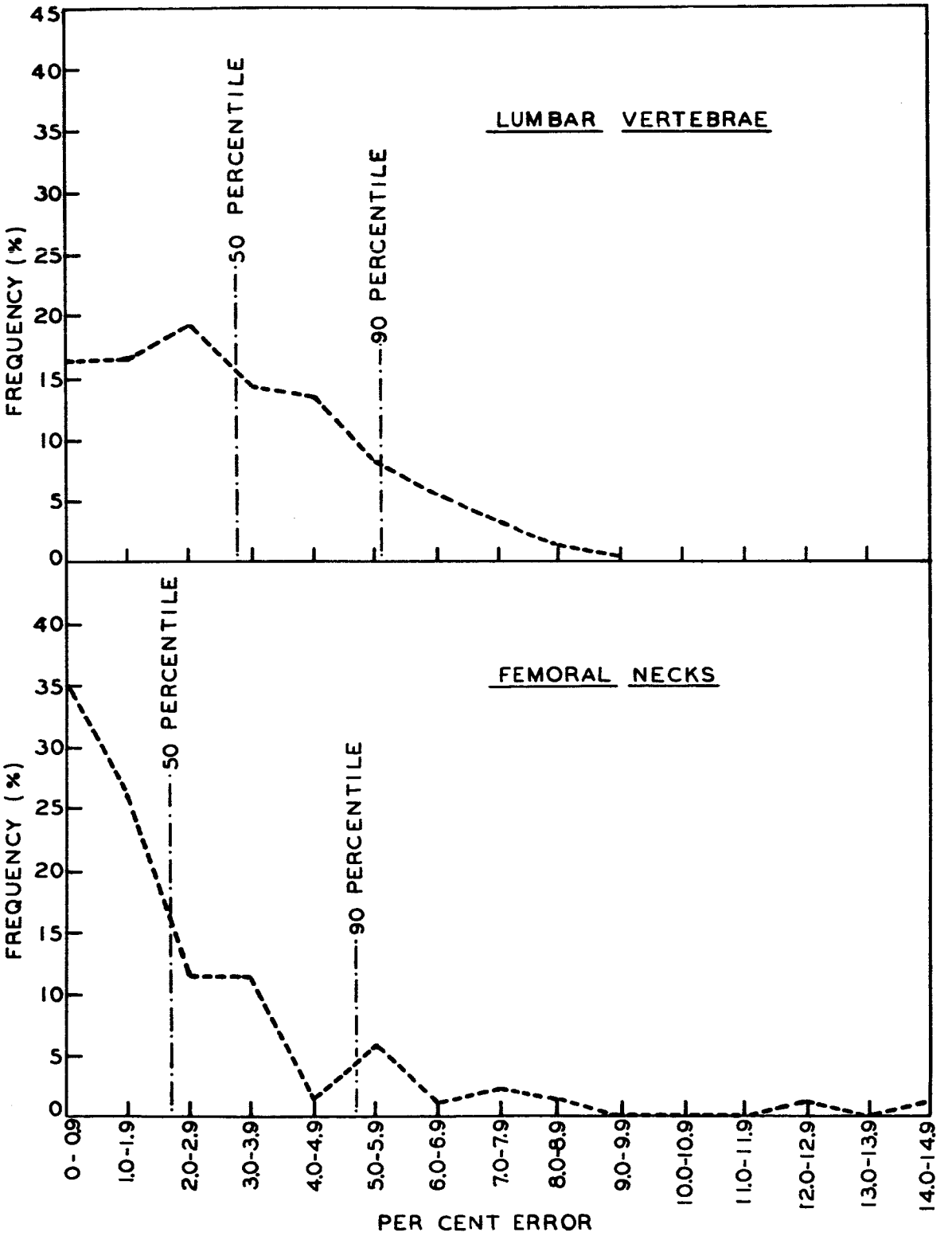

FIGURE 9.—Frequency distribution of errors between replicate evaluations of femoral necks and lumbar vertebrae.

error between replicate evaluations of femoral necks is somewhat less than that of vertebrae. In the case of the femoral neck, 50% of the time the error will be less than 2.9%, and 90% of the time it will be within 5.7%. In the case of the vertebrae, 50% of the time replicate values will agree within 2.9%, and 90% of the time the agreement will be within 6.0%

Since an average of two evaluations is taken for replicates within 2% for the femoral neck or within 3% for the vertebrae, it is probable that the error accountable to instrument and process reproducibility is less than 2%. Therefore, error arising from instrumentation or computation variations do not contribute materially to the overall error of the method.

CALCIUM CHANGES DETECTABLE BY RADIOGRAPHIC DENSITOMETRY

The value of any method of skeletal densitometry is, of course, its sensitivity in detecting small changes in bone mineralization rather than its high reproducibility alone. Within the current reproducibility limits of vertebral and femoral densitometry, the variations in X-ray aluminum equivalency accountable to experimental error have been interpreted in terms of equivalent calcium content on a mass basis in order to determine the magnitude by which calcium must be lost or gained in order to be detectable by the described method in its current state of development. In order for "X-ray aluminum equivalency" to have meaningful value in estimating ash content, the following anatomical or physical relationships must be known: (1) the relationship of X-ray aluminum equivalency to ash content (on a percent basis) in hydrated bone, (2) the mass of bone as estimated *in vivo*, and (3) the proportion of total ash composed of elemental calcium.

The first relationship has been measured during a study involving 105 cadavers X-rayed post-mortem, after which the body of vertebra L3 was removed at autopsy for ash analysis. The vertebral ash content on a weight-percent basis is determined by the formula 9.54+ (9.84 x aluminum equivalency) = percent ash, having a correlation coefficient of $r=0.93$.

The second relationship (that between bone mass and radiographic measurements) was determined during the same investigation in which the hydrated bone mass *in situ* was estimated by determining the relationship of bone dimensions to the weight of the fresh vertebral bodies removed at autopsy. It was found that the lateral cross-sectional area of L3 (cm^2), as determined from magnification-corrected lateral radiographs, was related to its hydrated mass (grams) by a factor of 0.049. The Standard Error of Estimate of vertebral mass was 4.2 g at the 68% confidence level and 8.4 g at the 95% confidence level. (The fully hydrated body of L3 in the adult male normally weighs about 50 g.) Although these errors obviously can be as large as 20%, they will not seriously affect the accuracy of determining calcium accretion since elemental calcium comprises only about 8% of the weight of fully hydrated bone, and secondly, because the same hydrated bone weight is used repeatedly in serial evaluations of the same individual.

The third factor, the proportion of bone ash comprised of calcium, is taken to be 0.4 which is the normal weight concentration of calcium in the hydroxyapatite molecule $Ca_{10}(PO_4)_6.(OH)_2$.

It should be emphasized here that in serial measurements of the same individual over a period of time, the aforementioned relationships, which are somewhat arbitrary, are not applied routinely. The simpler unit of "aluminum equivalency" permits the estimation of mineral gain or loss between consecutive radiographs, while the derived relationships have been used only to estimate the probable magnitude of the changes in milligrams of calcium.

Precision at the L3 Site

The lumbar spine of a male subject, age 21, was radiographed three times at 3-day intervals. The maximum difference among values of the three tests was 5% (aluminum equivalency range: 1.35–1.42 mm/cm). Bone dimensions scaled directly from the radiographs, subsequently corrected for image magnification, indicated a hydrated bone weight of 52 g. Such a

variation in aluminum equivalency corresponds to an apparent variation in vertebral ash content of 22.8 to 23.5% for an apparent maximum mass difference of 150 mg of calcium among the three determinations.

Surprisingly, these values determined at 3-day intervals are in somewhat closer agreement than three values made of a second male subject age 21, taken within a 10-minute period which yielded a precision of 170 mg of calcium. This indicates that the time variate did not significantly affect the result. Therefore, within the limits imposed presently by radiographic densitometry of the lumbar spine, changes of 170 mg of calcium within a single vertebral body (or 4.2% of the total calcium in the average adult male) appear to be the smallest amounts presently detectable by the method.

Precision at the Femoral Neck Site

Duplicate radiographs were made of the left femoral neck of a 21-year-old male on four testing dates at 5-day intervals. The tube, film, and subject were completely realigned for each exposure, and the four films yielded aluminum equivalency values agreeing within 2.5%, 2.7%, 0.6% and 1.3%, respectively. The largest variation would require, in theory, a change of 80 mg of calcium within the mid portion of the femoral neck (or 3.0% of the total calcium) to produce such a percent change in X-ray absorption. The 80-mg figure, therefore, is presently assumed to be the smallest change in calcium detectable within the bone section examined.

SUMMARY

A summary of probable errors accountable to the variable factors in radiographic densitometry is shown in table III. Although every attempt has been made to isolate the variable errors according to the categories listed, in some instances the total error will comprise two or more variable errors. For example, the 3.4% error in item no. 9, "repositioning of subject," is also affected by such variables as nonuniformity of radiographic emulsions and kilovoltage variations, although the darkroom processing factors and instrument errors are minimized.

TABLE III.—*Summary of Analyses of Variable Errors*

Factor	*Average error*
1. Instrument and process reproducibility.	2.0% (S.D.=1.5%)
2. Fluorescent screen sensitivity.	1.0% (S.D.=0.9%)
3. Intensity (max time) within 20% of optimum exposure.	0.5% (S.D.=0.2%)
4. Variations per kV change (58–62 kV):	
(a) Low MAS	10.2% (S.D.=2.0%)
(b) Intermediate MAS	5.2% (S.D.=2.2%)
(c) Moderately high MAS.	3.0% (S.D.=1.4%)
(d) High MAS	1.4% (S.D.=1.2%)
5. Temperature of developing solution	No measurable error between 67° and 69°
6. Age of developing solutions at constant temperature.	1.3% (one test only)
7. Routine darkroom errors when films processed at same time.	1.3% (S.D.=1.0%)
8. Routine darkroom errors when films processed at 24-hour intervals.	3.8% (S.D.=1.3%)
9. Repositioning of subject, L3 site.	2.1% (S.D.=1.1%)
10. Repositioning of subject, femoral neck site.	1.8% (S.D.=0.3%)
11. Different X-ray units (noncalibrated).	12.7% (S.D.=3.8)
12. Different X-ray units (calibrated against T.W.U. unit).	1.9% (S.D.=1.6%)
13. COMPOUND VARIABLE ERRORS MAY BE ACCOUNTABLE FOR TOTAL ERRORS OF UP TO 5% AT THE L3 SITE, AND UP TO 2.7% AT THE FEMORAL NECK SITE.	

Although kilovoltage variations may produce errors of 10% or more, at optimum exposure intensity the error is concluded to be 1.4% per kilovolt change.

The apparent error between two different X-ray units may be reduced from the maximum observed error of 31% to approximately 2% by use of an ionization chamber method of calibrating two units.

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The major error in X-ray densitometry of vertebra L3 and the femoral neck appears to be the failure to reposition the subject in precisely the same way in follow-up exposures. Currently, the total error appears to be approximately 2%, although errors of up to 5% may occur at the L3 site and up to 2.7% at the femoral neck site. Assuming the maximum error to be the case, the limits in determining calcium changes within a single bone between successive tests appear to be about 170 mg in vertebra L3 and 80 mg in the femoral neck. In adult males variations of this magnitude would indicate changes of 4.2% and 3.0% in vertebra L3 and the femoral neck, respectively, of the total calcium. It is possible that additional refinements can be made which will result in increased precision of the X-ray densitometric technique.

COMMENTS

Dr. CAMERON. Did you use grids in order to reduce scatter radiation?

Dr. VOSE. We had to use buttons.

Dr. CAMERON. It improves quite a bit. I think Dr. Rockoff's work was done without any grids, which also might affect the measurements.

Dr. VOSE. In addition, the phantom was lined in lead on the sides. The sleeve was slipped down over the phantom and we shot all pictures through an aperture in the lead sheet over the X-ray films, trying to cut out as much extraneous undercut scattering as we could.

Dr. SCHRAER. Did you have any results of vertebrae studies?

Dr. VOSE. Unfortunately, I do not have any photographs to show you, but it now appears, except for the gas which we can see on the X-ray film taken by our technique (and if we see it, we can eliminate it) that if we take another film the following day of the same patient, we appear to be working in not more than a 5% error. Duplicate films are made of each subject, and the films are evaluated twice. If they are found to be within 3%, then the average of the two values is taken.

Dr. RICH. What sort of a problem would be introduced if you were following a patient over some period of time and the patient lost weight? This would change the scatter. Could you compensate for this just by taking out some of your phantom?

Dr. VOSE. We would not dare to compensate for this by changing the kilovoltage even though we put the caliper across the patient. We would have to retain the same kilovoltage, and we just hope in the studies where this is applied that there won't be a tremendous loss of weight. If there were a tremendous loss of weight, we would have to retain the same kilovoltage, but possibly lower the intensity of exposure somewhat.

Dr. CAMERON. I am impressed with the results you get on these thick bones. I did not see the work on the vertebrae. I saw this mostly on the head of the femur. What does the vertebrae show?

Dr. VOSE. It is not quite as good as the neck of the femur. We feel that we can work within not more than 5% error in the third lumbar vertebrae, while we work within 3% on the neck of the femur.

Dr. SCHRAER. What about ash versus density value?

Dr. VOSE. With the help of Dr. Herster at Austin State Hospital, we evaluated cadavers to see the relationship of the X-ray value compared with the actual ash value of a bone at autopsy. We did the very difficult job of carrying the 106 bodies to the X-ray room, taping them up in a lateral position, and X-raying them. We then wheeled them off to the morgue and removed the third lumbar vertebrae. We plotted the values obtained densitometrically and found a correlation coefficient of 0.93.

There are various possible sources of error. If we had a spread of about 2% of the actual ash content on the curve, we think this could be improved. On a patient that has died of chronic illness, apparently there is more bowel gas than in a normal person. This was a complication in some instances, and caused some errors which seemed to indicate a 2% change in ash content.

Another source of possible error was that in removing the extraneous tissue from the bone, we had to maintain the bone at hydrated weight, and at the same time, remove all the tissue, leading to slight errors in obtaining the ash content. The ash content data were presented on a hydrated basis, not a dry ash basis. We had a higher correlation that way.

Dr. WHEDON. Dr. Vose, have you made some measurements of the reproducibility of the bone density measurement in the same individual from one time to another within a few days' or a few weeks' time?

Dr. VOSE. Yes. On one of the most recent tests, unfortunately, we found a slightly larger error when we X-rayed the same patients within 5 minutes than when we X-rayed him the following day. Nonetheless, we have tried to outguess this and estimate how much mineral must have been lost to account for such a change as we saw. In estimating the bone mass change *in vivo*, we decided that this change in the case of a femur neck would be about 70 mg of calcium, and in the case of the body of a third lumbar vertebra, it would be a maximum of 170 mg of calcium. That is the sensitivity seen at the present time.

Dr. WHEDON. What is that in terms of proportion to the total mass of the part by volume or weight?

Dr. VOSE. In the case of the vertebrae, this is the amount that would have to be lost from the entire vertebral body to be detected by this system.

In the case of the femur necks, the site we used has a volume of about 15 $cm.^3$ You have to lose that —70 mg of calcium in 15 cm^3 of femur neck. We

hope by some new techniques we are using to reduce this to approximately 20 mg.

Dr. WHEDON. Could you translate these weight figures into some sort of a percentile estimate?

Dr. NORDIN. There are about 6 g of ash in a vertebral body. Well, 160 mg of calcium is about 400 mg of ash, so that a loss of 400 mg in 6 g is about 7%.

Dr. SMITH. What is the geometric consideration that led you to pick that triangular area and its dimensions in the neck of the femur?

Dr. VOSE. This is what is called Ward's triangle, the part of the bone in osteoporosis that appeared to be demineralized visually before any other part of the femur neck. It is ideally located and is the part that is fractured with the transverse fracture of the femur neck. We thought it would be the most logical site for applying this measurement.

Dr. SMITH. How many individual density measurement readings were made within that triangle we saw?

Dr. VOSE. Four readings are routinely made.

Dr. SMITH. The variations may be high within your square.

Dr. VOSE. No, they are surprisingly close. We make four, but the average of the readings is very close.

Dr. LEROY. Dr. Vose, when you say that you can detect a loss of 6% from a vertical body, with what degree of confidence is this? At 95% confidence, or 90% confidence, or what?

Dr. VOSE. In the case of the third lumbar vertebra, the 90% confidence level for error is between about 5 and 5.9%; at the 50% level, it is 3.9%. In the case of the femoral neck, at the 90% confidence level, the percent error is 4 to 4.9%, and at the 70% level it is about 2.5%.

Dr. ROCKOFF. Dr. Vose, in your published graph of X-ray equivalency versus the vertebral ash content, the regression line on vertebral ash content goes through 8.5% ash and zero aluminum equivalency. That is, if the bone entirely disappears, according to your data, you still have 8.5% of vertebral ash present.

Dr. VOSE. We apparently do have a greater error on the bones that are very lowly mineralized. We did not dare curve that off, so we simply extended it and made a straight line. We do not seem to have enough points yet to determine just where the line should go. We do not dare predict the total calcium content, when the total bone ash is below 9 or 10%.

Dr. ROCKOFF. You have about ten points, as low as a tenth of aluminum equivalency, which extends out as far as 11 or 12% bone ash, which is large when you consider that 27% bone ash is the highest figure you have. I think there is a bias in the system.

Dr. VOSE. Obviously there should not be a straight slope all the way through that. We have also noticed that. In a few cases on the Herster's values we had to report a negative value because a straight line was shown. We do not think that is exactly right either.

Dr. NORDIN. Dr. Vose, did you exclude the gas filled corpses from that reject figure of yours?

Dr. VOSE. In some cases we did have to eliminate some of the cadavers. When we had a small gas pocket, we could visualize it by our system. I don't think you can visualize it by tomography.

Dr. NORDIN. Our studies on corpses has been with plain lumbar films, and I have found that the films are virtually unusable because of the combination of gas and feces.

Dr. VOSE. This is a more drastic problem than in living human beings, but if the gas pocket is large and if it is uniform, covering all of the body of the vertebrae and also the adjacent soft tissue, then it apparently cancels itself out somewhat.

Dr. RICH. The fact that you sometimes go down in these density studies—and other methods do this, too—to values that really look pretty much like or less than no bone at all may not be entirely due to these technical factors that I don't understand, because the vertebrae are filled with fat, of course. They can have a good deal of fat in the marrow. Fat has a lower mass coefficient that saline or water; therefore, in fact, the background in these measurements may not be the same as the same volume of muscle. In other words, the vertebrae that has almost no bone has something in there.

Dr. VOSE. We try to compensate for this soft tissue absorption by use of plastic, but in a few cases we have found vertebrae which had a 30% fat content.

Dr. RICH. And fat does have a lower mass coefficient?

Dr. VOSE. Fat has a lower mass coefficient than water or protein.

Dr. CAMERON. How easy would it be for a Department of Radiology in a hospital to do these measurements? Would it be a big project, do you think?

Dr. VOSE. One of the big problems is calibrating the X-ray machines. We have run test films on five X-ray machines in Denton and have obtained values with a maximum spread of 30%, using the same settings according to the kilovoltage meter. We have attempted to calibrate the machines, taking the worst pair; and by means of an ionization chamber, based in a lead boat under the tube, we determined the roentgens output on our own machine at the T.W.U., then experimented with one in the local hospital until we obtained the same energy output. This reduced the maximum 30% error to somewhat less than 2%, using the plastic phantom.

From one hospital to another this would definitely have to be done. We would have to calibrate the machines, one against another, assuming we had one central unit. The densitometry would definitely have to be done on one or two densitometers.

Dr. SCHRAER. In your graph, the title of the table is "Changes in Calcium Content of the Skeleton Detectable by Non-Destructive Techniques."

A subject is assumed to have 1000 grams of calcium in his skeleton. First, we will say that the method of detection is unknown for the time being, and the error in the technique, shall we say, is 1%. Now, 1% means

10 g of calcium in this case. If the calcium balance was 0.1 g per day, consistently up or down, this would mean that a hundred days would be required to see a 1% change, if this change was uniform over the whole skeleton. On a 0.4 g per day balance, this would require only 25 days. As the percent error in technique goes up to the 5% level, it would take a minimum of 125 days on 0.4 g per day balance to see a change.

Now for the conventional radiographic technique which most radiologists use (their eyes), this would require 750 days.

Clearly, then, any attempt at quantification is a big improvement over what the conventional radiologist can do. We cannot expect miracles in the first place. We cannot see changes as rapidly as we would like, but it is a better method by far than conventional radiography.

Dr. Vose. In addition, Dr. Schraer, we might mention that some bones apparently are more susceptible to change than others, and your calculations would be assuming that they are changing the same throughout the skeleton.

Dr. Whedon. Dr. Vose was just emphasizing a point that I think many of us would make: that loss of calcium under either pathological or physiological circumstances is not uniform across the skeleton. It is greater in one part of the skeleton than another. In large measure, of course, it is in proportion to the amount of trabecular bone that is available.

Dr. Rockoff. I would like to pursue the issue of linearization. I think linearization is an important issue, but not so much with techniques for measuring cortical thickness or with techniques which are looking for gross changes. If we are going to claim that we can identify minor changes, I think we have to define the validity of using this technique.

We all remember that the first approximation of the effect or, let us say, the change in radiation is given by the formula, i equals i zero e to the minus mu. Even when monochromatic radiation is used (the mu being some monochromatic amount of linear absorption coefficient of some monochromatic radiation) it is not a linear absorption function. This is further complicated by the fact, as I pointed out earlier, that with heterogeneous radiation there is a change in amount and then a change in frequency distribution.

Dr. Mack suggests that perhaps the straight line relationship (fig. 12, "counts" vs. stepwise increases in calcium) is the correct one because it is a straight line, and I point out that it is a straight line because the relationship was made a straight line relationship by the linearization technique.

Dr. Vogt. Nature gave us this nonlinear curve with the wedge that we get out of the first recorder, but Nature also gave us a linear wedge of which we took a film at the same time that we X-rayed bone or calcium.

We can convert a nonlinear function into a linear function or to provide a calibration factor, and that is what we are doing. This is a calibration technique rather than a straightening out of a trace which means something else.

Dr. Mack's mechanical and electrical system is an integrating microdensitometer. Put in simpler words, it is an electromechanical servo system, probably one of the simplest servo systems that exist.

The initial steps for preparing a film to be put into the densitometer consist, first of all, in the procedures involved in the technique of taking the film, in the placement of the subject, in the placement of the film and wedge, the placement of the X-ray beam, which is set by means of a light beam, and all the way through the developing technique, the handling of the films prior to processing, and locating the two anatomical landmarks that you are going to scan.

Any time you discuss a technique in which you are getting good reproducibility, you have to discuss the method of acquiring the data as well as the instrumentation that you used to record it.

The primary purpose of this instrumentation system is to make every wedge on every film equivalent to one another. In other words, as Dr. Rockoff pointed out this morning, there are many factors that can influence the optical density of an X-ray film, if you don't make corrections or at least do the same thing to the film each time. Even if you do the same thing to the film each time, you can get different optical densities. Therefore, as has been pointed out, we need some reference.

Dr. Mack starts out by having no light strike her detector. This is zero transmission of light to the detector. She then goes out 13 cm on the wedge (it is the same wedge on every film) and gets a certain amount of light transmission. If this X-ray film had been exposed differently from a preceding one, she might get a different optical density. In fact, she would get a different light transmission. Therefore, some correction has to be made to make this wedge equivalent to the wedge on the previous film. This is done by adjusting the light source through the film so that 13 cm out the wedge gives full scale deflection on the recorder. Her first recorder initially is set up so that zero reading is zero light transmission, and 100% or full scale reading is the transmission of light 13 cm out the wedge. In other words, we are making every wedge on every film the same.

Then as Dr. Mack traces this first film, she gets a nonlinear type of trace, as you saw for the 5:1 wedge and the 10:1 wedge, its nonlinearity depending upon what portion of the full H and D curve you are on. Because of this nonlinear trace, there must be some means to convert it or to make a multiplication factor for reach reading. This is done by means of a slidewire potentiometer. This now becomes the electromechanical part because it is mechanically coupled to the pin on the recorder. A correction is made along each of the 20 sections to make the raw film give a reading that you

would expect for the aluminum which you know to be a linear function.

Dr. Mack has a technique that has been developed by reading from the raw curve with a little scale that is calibrated. She can set the 20 potentiometers which are shunted with a 50-ohms potentiometer which has a thousand steps in it, providing a resistance range of from zero to 33 ohms. Since this is a mechanical system, linked directly, and the recorder is driven by direct coupling, any correction made at one part of this curve does not influence the correction made at any other part of the curve. In actual practice, when Dr. Mack is making a densitometry tracing and calibrates the wedge (or if you like, linearizes the wedge), the wedge is linear. Therefore it does not matter whether one goes from zero to the full range or simply linearizes it over a portion.

I feel that since the range of bone density measurements made on the groups of subjects we worked with, going from about 50 to around 80% of the total wedge length, does not make any difference, it is good to have the wedge as she has it.

Now again, we get away from the electrical part of the system and come back into the mechanical part of the system. This integrator is mechanically linked to the pin on the recorder so that the displacement or drive of the pin from zero gives a certain displacement of the slidewire potentiometer, which then drives the integrator, and integrates the area under the second curve.

To show the reproducibility of densitometry of the os calcis, eight films were taken of my left os calcis at a hundred million amperes, 60 kV, and 0.6 second exposure. I got on and off the table between each of the eight films, was repositioned, and an exposure was made.

After the films were developed, each was run through the densitometer three times, and the data show the reproducibility. The statistician reports that the mean total for the total scan across the os calcis is 11,092.79, ±75.12. At the 99% confidence level, this presents an error or standard error of ±0.75% and if we add them together, we get 1½%.

Dr. ZELEN. That is standard error?

Dr. VOGT. Right.

Dr. ZELEN. What you really want to quote is the variation between films, that is, what agreements you would expect to get on two independent measurements by two different films.

Dr. VOGT. I have these here, too, in much more detail, but I think this really shows the same thing. I realize that the more times you have, the better you get, but at least it tells you for your 99% confidence level what range or reproducibility or error that you are talking about.

Dr. ZELEN. It is an inappropriate figure to give.

Dr. WHEDON. Standard error has more to do with the validity of the mean than it has to do with indicating what the variation is. An appropriate figure to give for reproducibility is the standard deviation.

[*Editor's postconference note:* Review of Dr. Mack's table I with Dr. J. Z. Hearon, Office of Mathematical Research, NIAMD (NIH), indicates that the mean for the 24 total integrator counts columns is 11,160, with a standard deviation of ±51.4; although the standard error was not actually calculated, it would be less, approximately 20. The meaning of this is that, even though Dr. Vogt's figures are not intelligible to us, he and his statistician have been very conservative and that for repeated films at the same time on the same individual, the method appears to be *highly* reproducible. An additional observation of particular pertinence to practical use of the method would be to take one film on the same individual on several successive days, then calculate the standard deviation of this group of measurements.—D. W.]

Dr. LUTWAK. It would help to solve the controversy in my mind if you were to take the calcium data, calibrated from the linearized system, and put them back through the system again, backward; would you then come up with the HD shaped curve? If you did, then I do not think there is any controversy.

Dr. VOGT. Certainly.

Dr. LUTWAK. I think the problem is one of semantics again. We are talking about linearization. You are really not linearizing; you are calibrating, and you are establishing the standard curve which is easier for the machine to handle and for you to see.

Dr. VOGT. For the special purpose an analog computer is used.

Dr. MACK. If you were doing something where your values would be quite low, you would not use the simple linearization wedge; you would not use the same wedge that you used where the values are scattered. We pick our wedges according to the bone to be measured: for the little finger, we have a different one from that for the os calcis, and still a different wedge for the back.

Comparison of Cortical Thickness and Radiographic Microdensitometry in the Measurement of Bone Loss

STANLEY M. GARN, ELISE FEUTZ, CHARLES COLBERT, AND BETTY WAGNER, Fels Research Institute

In contrast to radiographic microdensitometry, which has the longer history of application despite greater technological complexity, the far simpler measurement of cortical thickness on radiographs (i.e., microradiogrammetry) is virtually a technical newcomer. Prior to 1960 very few attempts to employ cortical thickness were made; and, except for Trotter's pioneering comparison of cortical thickness to absolute bone volume in the femur (Trotter, 1954), these attempts were largely restricted to studies primarily of genetical import (Hewitt, 1957/8; Garn, 1962).

Since Virtama and Mähönen (1960), there has been a considerable and increasing interest in the measurement of cortical thickness on X-rays. A growing volume of reports have come from Nordin and his group (1960, 1965). Meema and his associates (Meema, 1963; Meema and Meema, 1963), Smith and his associates (Smith and Walker, 1964; Smith and Frame, 1965; Smith, 1965), and the present author and his associates (Garn et al., 1963, 1964; Garn et al., 1964; Garn et al., 1964–a, b, c, d; Garn et al, 1965).

The measurement of cortical thickness directly on radiographs represents a novel and important approach, obviously far more suitable for tubular bones than for purely spongy bones, a substitute for conventional radiographic microdensitometry in some cases, and a replacement in others.

RADIOGRAPHIC TECHNIQUES

Radiographic techniques are not critical for the measurement of cortical thickness. Either nonscreen or screen-type film can be employed, though the former gives superior detail. Hospital, clinic, and survey films can be used for this purpose. The PKV–MAS combination is not critical, though high PKV techniques (90–120 PKV) give flatter results for the hand and foot; and the 30–50 PKV range is greatly preferred. Small variations in the tube-to-film distance are not critical, though a completely fixed tube-to-film distance in meauring short term rates of bone loss is used. Coning-down and processing variables do not introduce errors in measurement, as they do in "bone densitometry." The same films can be used for the measurement of cortical thickness and for conventional microdensitometry, in which case nonscreen film, a rather heavy exposure, and careful attention to processing and coning-down are then mandatory.

MEASURING TECHNIQUES

Measuring equipment is not a major problem in making cortical thickness measurements. Much of our work over the last 5 years has been done with a stock $7.00 Fisher catalog Vernier caliper with 0.1 mm readout, modified by grinding the tips. We now prefer the $70.00 dial-reading, No. 1068C 6-inch replaceable needle-tip Helios metric caliper with 0.05 mm

readout (Carl Neise, New York); and for some purposes we have employed the 0.1 mm readout, large capacity, automatic print-out GOAT, manufactured by the Gerber Instrument Company (Hartford, Conn.). This latter instrument prints the measurements directly onto paper tape; and, as it is also motor driven, it obviates both caliper fatigue and error due to incorrect reading of Vernier or dial calibrations.

PERSONNEL FACTORS

In studying cortical thickness, technician training has not been a problem. Though our preference has been for biologically trained college undergraduates suitably interested and motivated, there is nothing about the technique itself that places unique demands upon the intelligent technician with some knowledge of bone anatomy and familiarity with radiographic techniques.

REPLICABILITY AND VALIDITY OF CORTICAL THICKNESS MEASUREMENTS

Replicability of the cortical thickness measurements is high, being of the order of 0.97–0.99 both for intraobserver and interobserver replicability. Long term intraobserver replicability is high, employing radiographs of the same individual taken many years apart. In a recent study comparing loss of compact bone over a 15 to 30-year period in the same individuals, we were able to repeat earlier estimates of inter- and intraobserver reliability for the same subjects. Although there is slightly greater difficulty in ascertaining the endosteal surface in older and osteoporotic individuals, interobserver and intraobserver reliability was of the same order of magnitude in each case (see table 1). Using the Helios caliper with 0.05 mm readout, it was shown that the RMS readout error was 0.15 mm. Since the average adult cortical thickness is between 4 and 7 mm in the second metacarpal, as Smith, Nordin, and our group have shown, it should be possible to show a decrease of the order of 5% or less with considerable reliability.

There is the question of where to measure the cortical thickness on bones such as the second metacarpal that are effectively dumbbell or spool-shaped. We have taken the cortical measurement at mid-shaft, an arbitrary point, but one which simplifies the speed of readout and maximizes reliability. Dr. Richmond Smith and his group make the cortical thickness measurement at the area of minimum medullary (marrow) cavity width, a point which may be considered to have greater anatomical meaning. There appears to be very good agreement between the two approaches, as shown by the comparison of our measurements and those of Dr. Richmond Smith. The mid-shaft measurement appears simpler for routine assessment and more replicable (fig. 1). There is no absolute basis to prefer one measurement technique over the other.

We have explored the validity of the cortical thickness measurements, employing the measurements of cortical thickness on the radiographs with pinpoint calipers with the measurements of cortical thickness made from microdensitometric traces of the same bones (see fig.

TABLE I.—*Inter-Observer and Intra-Observer Replicability of Cortical Bone Measurements* [a]

Subjects	Type of replicability	*N*	*r*
Fels subjects	Intra-observer (PN)	20	0.98
Fels subjects	Intra-observer (DA)	20	.95
Fels subjects	Inter-observer (PN–DA)	20	>.99
Fels subjects	Inter-observer (PN–DA)	20	>.99
Fels mothers (1945)	Inter-observer (EH–ML)	43	.98
Fels mothers (1965)	Inter-observer (EH–ML)	43	.99
Senior citizens	Inter-observer (EH–ML)	25	.98

[a] Second metacarpal at mid-shaft.

2). Considering the theoretical bone model as that of a simple cylinder and referring to radiographs of this theoretical bone model simulated by aluminum tubing, the true amount of "cortex" can be understood, and the radiographs interpreted in terms of the theoretical bone model. Comparing cortex by micrometry with that taken from the densitometric "trace," the caliper measurement taken directly on the X-rays appear to be highly correlated with the trace measurement; however they systematically overestimate the true cortical diameter (table II). The caliper measurements subtract the bone medullary cavity from the total periosteal diameter, slightly exaggerating the latter (i.e., measuring just outside of the true periosteal diameter) and slightly underestimating the medullary cavity (i.e., measuring just inside of the endosteal margin). It has been suggested that superior results might be obtained by measuring the enlarged image of the radiographs instead of working directly on the

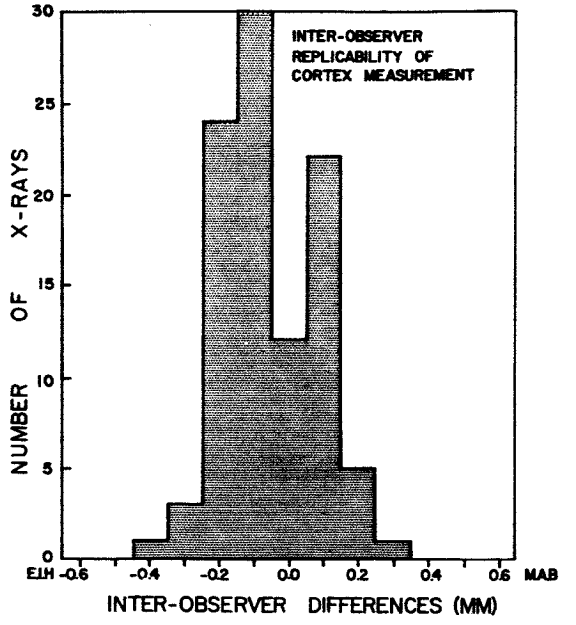

FIGURE 1.—Interobserver differences in cortical thickness of the second metacarpal at mid-shaft as measured on 98 radiographs of adult females. As shown, the RMS error is of the order of 0.15 mm. With intra- and interobserver replicability better than 0.98, and with interobserver differences of less than 0.2 mm, bone loss of the order of 5% may be reliably measured for the bone in question.

TABLE II.—*Validation of Cortical Thickness Measurements*

(Comparison of caliper micrometry with measurements taken from the densitometric trace[1])

Group	N	r
1. St. Louis Negro males	52	0.87
2. St. Louis Negro females	97	0.93
3. Combined sex	149	0.93

[1] Cortical thickness by micrometry by Philip Nolan, Jr., and cortical thickness measurements from Joyce-Loebl densitometric trace by Elise Feutz.

radiographic images themselves. We have attempted this, but the fall-off of definition at even moderate enlargement (5–10 times) complicates rather than improves the situation (see also fig. 3).

In the process of validation there is the comparison of the cortical diameters, as measured above, with the aluminum equivalent densities taken directly from the microdensitometric traces made at the same point. A large series of Negro second metacarpals were X-rayed at Washington University School of Medicine by Christabel G. Rohmann. The radiographs were returned to the Fels Research Institute for both caliper measurement and densitometric measurement. The measurements of cortical thickness taken on the medial and lateral aspects of the X-rays do not correspond exactly to the thickness of the cortex directly in the optical path (i.e., the thickness of the cortex *wall* on the dorsal and ventral sides of the bone). This comparison was made because cortical thickness on the second metacarpal *must* be taken as described above *in vivo*, while the densitometric measurement must reflect the amount of cortex *in* the optical path, since bones in the living subject are not free for separate measurement in both the antero-posterior and lateral views! This comparison of cortical thickness and radiographic density assumes that the bulk of the radiographic density in the second metacarpal is confined to the cortex *per se*, an assumption which we have clarified by taking radiographs of isolated metacarpal bones, cutting them in two at the mid-section, removing bone in the medullary region, and

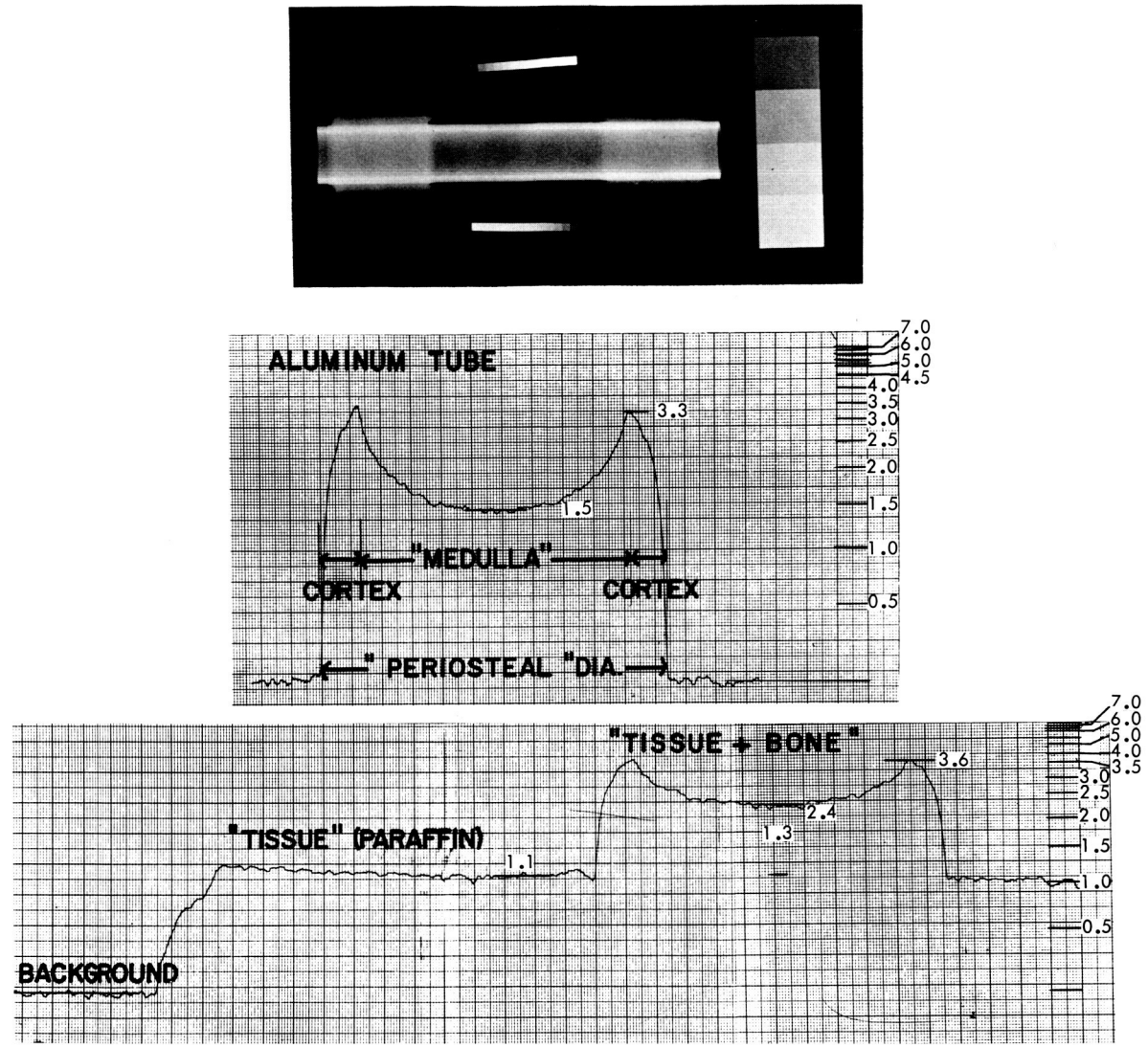

FIGURE 2.—*Above:* Radiograph of aluminum tube used as a simulated tubular bone model. By making densitometric traces on the radiographic image of this tube with and without simulated soft tissue, the meaning of the densitometric trace becomes clearer. *Below:* Microdensitometric traces of simulated bone and bone-plus-tissue radiographs. Configurations of the "cortex" and "medulla" are clearly seen. It is possible to interpret actual Joyce-Loebl microdensitometric traces of small tubular bones.

then again subjecting them to radiography and densitometric examinations (see fig. 4). The obtained correlation of 0.6 between the thickness of the medial and lateral cortical walls, as measured by caliper, and the aluminum equivalent density, as measured from the same radiographs, represents a reasonable degree of agreement. The true correlation (correcting for attenuation of each of the measurements), the departure from circularity of the bones involved, and the lack of concentricity of the medullary cavity may be estimated as of the order of 0.85 or better.

We can effectively describe the cortical thickness measurements as (a) having excellent replicability and (b) good validity, though it is clear that measuring thickness of cortex on each side of the bone at mid-shaft is not identical with the measurement of radiographic density at the middle of the same bone.

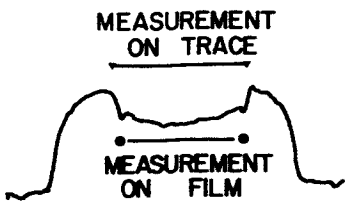

FIGURE 3.—Comparison of measurement of the medullary cavity by micrometer directly on the film and from the microdensitometric trace. Since the medullary cavity measurement by micrometer tends to be slightly smaller than the true value, the apparent cortical thickness tends to be systematically exaggerated as compared to measurements made on the microdensitometric trace. There is justification, therefore, for using the microdensitometric trace in place of on-film caliper micrometry.

REPORTING CORTICAL THICKNESS

In discussing the various advantages and disadvantages of the cortical thickness measurement, there is the important question of how to best report it. The averaged cortical thickness (L+R/2) corresponds most nearly to the wall thickness of the theoretical tubular bone model. The summed cortical thickness (L+R) corresponds most nearly to the aluminum equivalent density measured at mid-shaft. A cortical *area* estimate (i.e., the total cross-sectional area minus the marrow area) should correspond to the total bone material under the trace and, indirectly, to Frost's Absolute Bone Volume. Trotter's comparison of cortical thicknesses on the femur to the total bone weight (Trotter, 1954) and our comparison of estimated cortical volume to Garrow's measurements of bone mineral (Garn et al., 1965) reflect the extent to which the periosteal diameter, or total diameter, and the cortical thickness together reflect the amount of bone that is there.

Various workers have expressed these raw dimensions in various ways. Under the assumption that the cortical thickness represents the wall thickness and is the only measurement subject to change in adults, we have reported cortical thickness alone. Nordin (1960, 1965) and others express the cortex as a ratio, as a percentage of the total periosteal diameter. Some workers have expressed cortical area as a percentage of the total periosteal area. With age, all of these measurements decrease except total periosteal diameter and all ratios involving the cortex decrease. Periosteal diameter does not change appreciably after adulthood. While changes in the cortical diameter and the marrow cavity are necessarily reciprocal, it is the cortex that changes, and all measurements or ratios derived from the cortex are "derivative."

The averaged cortical thickness (L+R/2) and the summed cortical thickness (L+R) are the same measurement. The cortex-to-total ratio is heavily weighted by cortex, and the cortical area to total area ratio is primarily weighted by cortical area. We have compared various measurements and ratios involving the cortex as shown in table III. The correlations among the various raw measurements and ratios confirm the expectation that ratios containing the same values as numerator or denominator are heavily correlated, the correlations in this case meaning that they effectively say the same thing.

TABLE III.—*Comparison of Methods of Expressing the Bony Cortex Measurement (in 61 white males aged 25.0–34.9)*

	Cortical thickness	Cortical ratio	Cortical area	Cortical area ratio
Cortical thickness [a]	------	0.76	0.40	0.64
Cortical ratio [b]	------	------	−.12	.88
Cortical area	------	------	------	.11
Cortical area ratio [c]	------	------	------	------

[a] Sum of left and right sides.
[b] Cortical diameter/periosteal diameter.
[c] Cortical area/periosteal area.

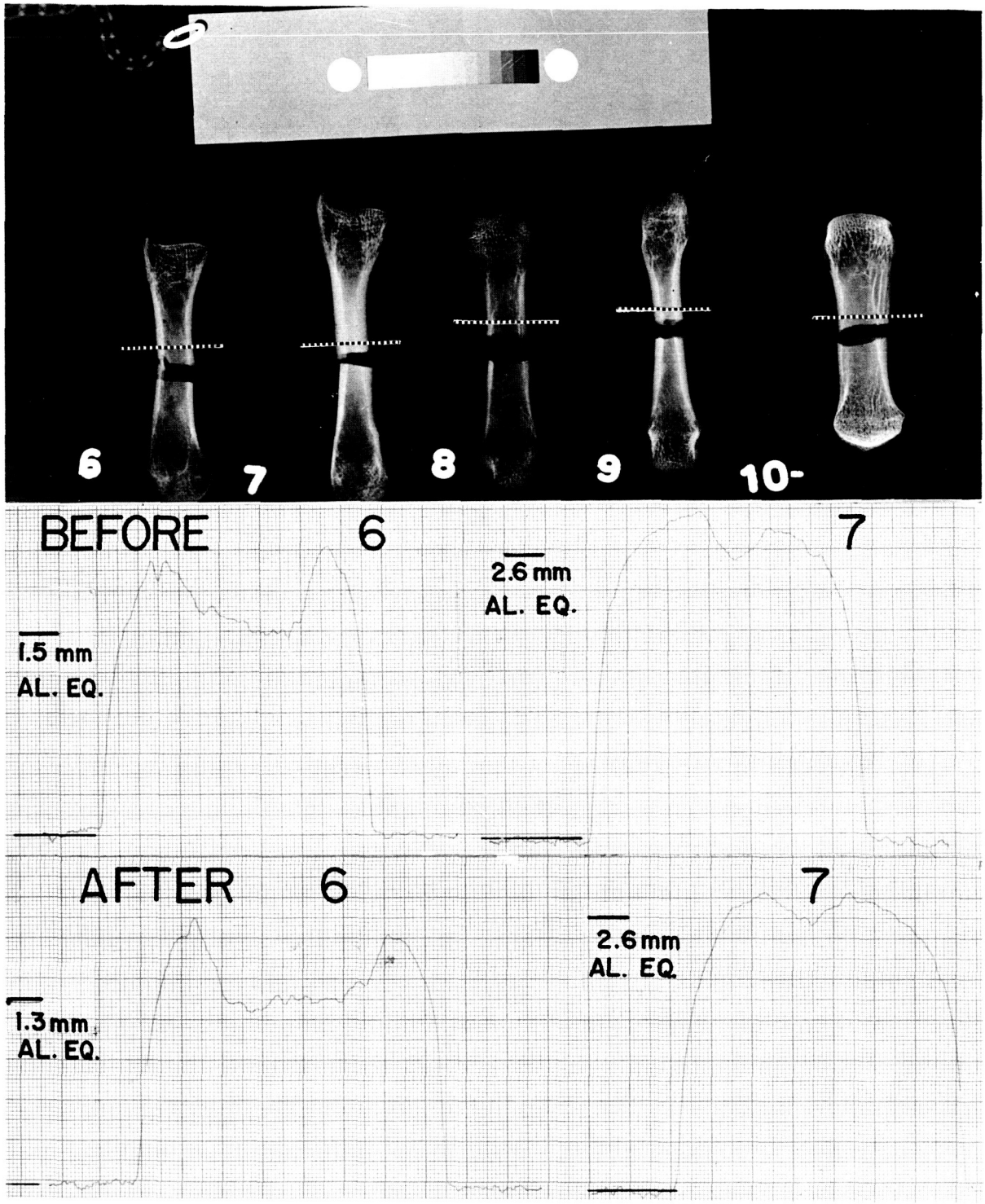

FIGURE 4.—Evidence that the radiographic density of metacarpals primarily resides in the cortex of the shaft. For a series of 10 metacarpals, 5 of which are shown here, "before" and "after" densitometric traces are substantially identical, despite removal of medullary material in the proximal half of each shaft. Densitometric traces of metacarpal #6 and #7, made along the paths indicated by the dotted lines, show no more difference than can be attributed to slight differences in repositioning. Accordingly, the cortex contains the preponderance of radio-opaque material in the metacarpals at mid-shaft.

We continue to favor the averaged or summed cortical thickness. The cortex is the active parameter and the parameter that changes in osteoporotic bone loss in tubular bone. There would be justification for reporting the cortical area corresponding to the Absolute Bone volume; and this may be desirable in tubular bones that approach the theoretical bone model in being (a) circular, (b) concentric, etc. The metacarpals are more or less circular, the width being 116% to 130% of their depth at mid-shaft. For other bones, whose circularity is not certain, as in most tubular bones, or where the antero-posterior view alone loses some of the bone materials (as in the femur), the computed cortical area does not add additional information. In the case of the femur, where the large pilaster of the linear aspera contains much bone and yet is not reflected in the thickness of the lateral and medical cortex, cortical area computed from the antero-posterior view alone is erroneous.

Relating cortical thickness to the total periosteal bone diameter is useful only if there is a relationship between the two. We might expect the bigger bone with the larger periosteal diameter to have a thicker cortex. In children and subadults, individuals with the larger periosteal diameter generally have greater cortical thickness as well. In adults the relationship between total diameter and cortical thickness within an age group and for a given sex and a single population is far from high. We prefer to measure and report cortical thickness *per se* since it best reflects the actual bone. Other expressions, including cortex as the percentage of total, or cortical area as a percentage of total area, will be highly correlated with cortex except that derivative ratios or ratios of areas then introduce errors and uncertainties, especially as the bone section departs from circularity.

DISADVANTAGES OF CORTICAL THICKNESS

As with radiographic densitometry, where fall-off of density from the center introduces errors of a major order of magnitude, and where processing variables (including streaking), fog, and soft tissue correction introduce multiplicative problems that have not been fully resolved, cortical thickness measurements have their limitations. Cortical thickness measurements cannot be applied to bones that are primarily spongy. Some of the finger bones fall in this category, the cortical portion of the morphologically variable middle segment of the fifth digit being very small and highly variable. Also the weight-bearing tubular first metatarsal bone falls in the latter group; there is too little cortex remaining to measure in older subjects.

Positioning may constitute a problem. It is difficult to malpostion the postero-anterior foot or hand views unless an oblique view or a rotated view has been intentionally ordered, and although a postero-anterior lower leg view may be reasonably "standard," variability in positioning may be too great for reliable measurement of other body parts. Medial rotation or lateral rotation of the lower leg can make successive X-rays of the same subject uncomparable. In analyzing bone loss in fracture cases seen at the Springfield City Hospital this year, replicability in positioning has been a most difficult technical problem. The immersion-positioning procedures for the forearm, used by Meema (1963) and Meema and Meema (1963), or the standardized forearm views employed at the Fels Research Institute are recommended. The less a bone approaches a simple cylinder in cross-section, the more positioning error contributes to unsatisfactory results.

The cortical thickness measurement may account for only a part (the greater part) of the mineral content of the bone. Some part of the mineral of the bone resides in the spongy center. In the metacarpals, the cortical bone (with its very high physical and radiographic density) ordinarily accounts for more than 90% of the bone mineral in the bone. This appears to be true of the femur at mid-shaft as well. The extent to which the cortical area of any bone represents the major portion of the bone mineral may easily be ascertained by the empir-

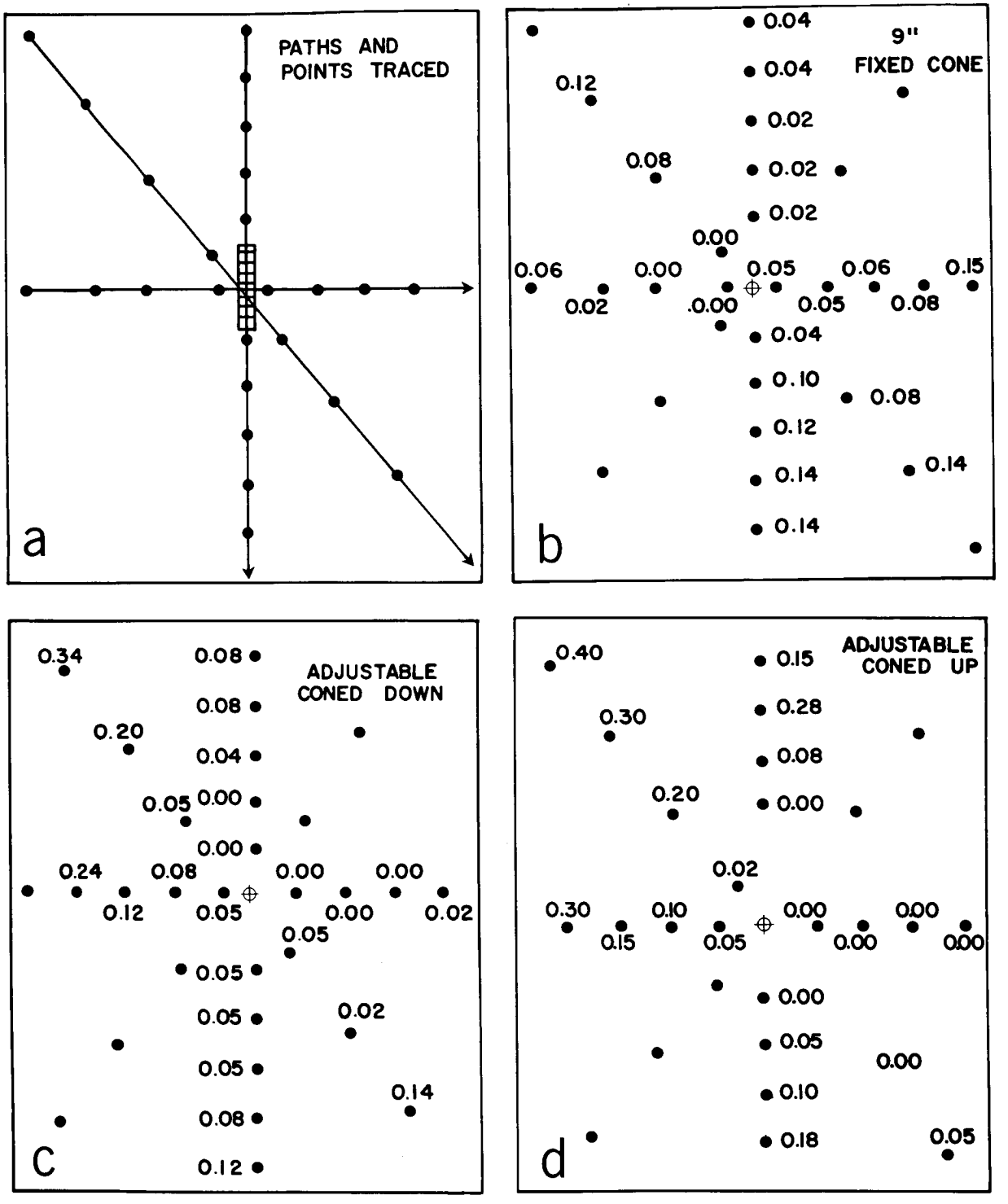

FIGURE 5.—Fall-off density from the film center shown for different cones and degrees of coning-down. Here, comparing a 9-inch fixed cone and a large adjustable rectangular "Collimator" cone, it is obvious that fall-off of radiation from the central spot constitutes a major potential source of error in radiographic microdensitometry, with aluminum-equivalent density gradients as high as 0.40 mm (corresponding to as much as 0.80 mm of cortex). Under these circumstances it is most inadvisable to make densitometric comparisons of radiographic films produced under a variety of field and clinical situations and it is most desirable to keep the reference wedge within a short distance of the bone site to be traced.

ical approach we have employed, using dry bones, or by comparing cortical thickness measurements and density traces of the same bone.

ERRORS IN THE DENSITOMETRIC APPROACH

Errors in cortical thickness measurement are of two distinct kinds: random, as exemplified by the RMS measuring error, and systematic, as shown by the comparison of measurements taken directly from the densitometric trace with those obtained by caliper micrometry of the same tubular bones. Systematic, also, are errors due to over- or under-estimation of the true cortical area when only a single radiographic view is used. Random errors amount to 2%–5% for the second metacarpal and may be proportionately smaller for larger bones with thicker cortices. Systematic errors may be ignored in certain of the applications or eliminated by not making area estimates from inadequate data. Measured today and remeasured tomorrow, a cortical width of 5.5 mm will be 5.5 mm $\pm$ 0.15 mm.

For the same bone, the second metacarpal, and for comparable radiographs, errors in the densitometric approach are of several kinds, occasionally additive and in some cases of major magnitude. One source of error is derived from the falling-off of the X-ray beam density from the central ray, a phenomenon due in part to the cone and the degree of coning-down. We have studied this error extensively and find it to be equal to as much as 4.4 mm of aluminum at 10.0 cm from the central ray (see fig. 5). Expressing this error as an RMS error of approximately 0.2 mm of aluminum equal to approximately 0.4 mm of cortex, it may be of the order of $\pm$10% in replicate measurements in the same second metacarpal and 20% or more in comparison to the aluminum equivalent thickness of the middle segment of the fifth digit. Visiting Meema's laboratory, we find that he has attempted to minimize this source of error by limiting the distance between reference wedge and the part being measured, knowing beforehand the extent of fall-off of radiation under the circumstances of his study.

A second error comes from processing streaks, due to insufficient agitation, close proximity between adjacent films, streaking from film holders, etc. Such areas of reduced density may be approximately equal to 0.4 to 0.5 mm of aluminum, or approximately 1.0 mm of cortex. We have attempted to minimize errors due to processing variables of all kinds by the use of nitrogen-burst processing (Calumet Manufacturing Company, Chicago). Processing streaks from all sources constitute a source of error. To some extent they can be "edited" out when they appear in areas of maximum density. When processing density variations occur in the useful area of the film (i.e., manus or pes), they may not be noticed.

A third source of densitometric error arises from the useful limits of a film-wedge combination; that is, when the optical density of the most radioopaque area of the wedge or body part comes close to the minimum optical density of the film itself. Minimum densities of processed Kodak nonscreen film approximate 0.3, though they may be as high as 0.5. About 0.05 of the optical density is due to the film base itself, 0.05 to the blue dye, 0.10 to processing, and 0.10 to scatter. The practical effects of this are shown graphically in figure 6. As the minimum density of the image of the wedge approaches the residual fog level, the log-linear relationship between aluminum equivalent thickness and optical density is altered. The calibration curve rolls off at this point and any estimation of an aluminum equivalent value of bone "density" would be in error, in some cases overestimating the true aluminum equivalent thickness by a substantial amount. The remedy for this source of error is, first, technically maintaining heavy enough exposures to pull the minimum wedge density above fog level; and, second, to correct for it where this is feasible, as shown in the figure.

Considering three sources of error, (1) that due to fall-off density, (2) that due to processing streaks, and (3) that due to calibration curve curve roll-off, it should be clear why the RMS error per aluminum equivalent thickness of small tubular bones may be larger, far larger than for simple measurement of cortical thick-

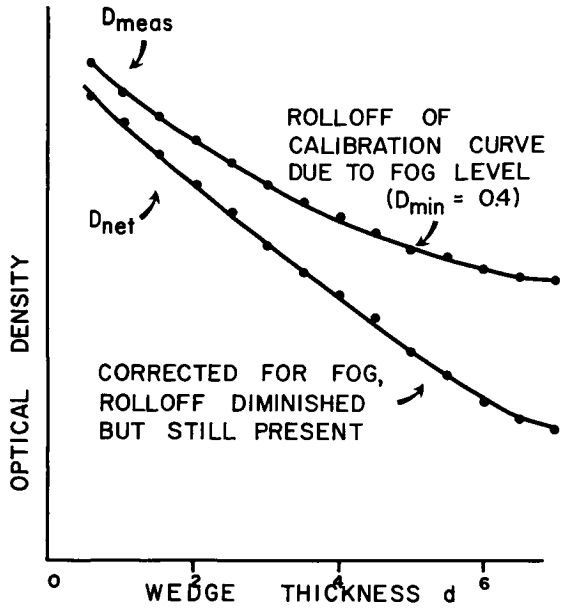

FIGURE 6.—Showing roll-off calibration curve as the minimum optical density of the wedge approaches fog density. Where the useful limits of the film-wedge combination are exceeded, estimates of aluminum equivalent density may be seriously in error, contributing major errors to radiogrammetric microdensitometry.

ness. They may be additive. While such errors can be edited out to some extent by preliminary quality control, they remain indeterminate to some extent.

SUMMARY

Study of many thousands of individuals (infants, children, adults, and octogenarians), has shown that measurement of cortical thickness is technically simple. It can be applied to previously taken radiographs, and it is particularly applicable to studies involving bone change. A loss of as little as 5% of cortical thickness may be measured reliably with total cortical material exceeding 5.0 mm.

The absolute reliability of cortical measurement is high (better than 0.98), the validity is good (exceeding 0.90), and the cortical measurement effectively measures that which changes most in tubular bone, i.e., the cortex.

Cortical thickness measurements demand minimal deviation from the usual radiographic procedures. Neither type nor processing of film are critical, and the equipment for cortical thickness measurement is simple and inexpensive. For those bones that approach the theoretical tubular bone model, cortical thickness measurements effectively and quantitatively describe how much bone is there and how much has been lost. Since the aluminum equivalent thickness per millimeter of young cortex and old cortex appear to be the same, the meaning of cortical thickness is both simple and direct.

Cortical thickness measurements are not affected by fall-off of illumination from the central ray, processing streaks or other processing errors, or deformation of the calibration curve—all of which can make for substantial errors in densitometric assessment. However, for spongy bones, including the vertebrae, for tubular bones with little cortical material such as the first metatarsal, and for flat bones such as the mandible, a densitometric approach remains inescapable. Densitometric trace can, in many instances, yield the same information on size obtained by micrometry; and with due attention to the anatomical meaning of the bone trace, qualitative as well as quantitative information on the sites of the bone loss may be obtained.

ACKNOWLEDGMENTS

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REFERENCES

BARNETT, E.; and NORDIN, B. E. C.: The Radiological Diagnosis of Osteoporosis. Clin. Radiol., vol. 11, 1960, pp. 166–174.

BÉHAR, M.; ROHMANN, C.; WILSON, D.; VITERI, F.; and GARN, S. M.: Osseous Development in Children with Kwashiorkor. Federation Proc., vol. 24, 1964, p. 338.

GARN, S. M.: The Genetics of Normal Human Growth. In: De Genetica Medica, Luigi, ed. Gregor Mendel Institute, Rome, 1962.

GARN, S. M.; ROHMANN, C. G.; and NOLAN, P., Jr.: Studies on the Development of Compact Bone in Normal Individuals and in Endocrine and Nutritional Abnormalities. Department of Growth and Genetics, Fels Research Institute, Yellow Springs, Ohio. Privately printed, 1963.

GARN, S. M.; PAO, E. M.; and RIHL, M. E.: Compact Bone in Chinese and Japanese. Science, vol. 143, 1964, pp. 1439–1440.

GARN, S. M.; ROHMANN, C. G.; and NOLAN, P., Jr.: The Developmental Nature of Bone Changes During Aging. In: Relations of Development and Aging, James E. Birren, ed. Charles C. Thomas, Springfield, Ill., 1964.b.

GARN, S. M.: BÉHAR, M.; ROHMANN, C.; VITERI, F.; and WILSON, D.: Catch-up Bone Develepoment During Treatment of Kwashiorkor. Federation Proc., vol. 24, 1964, p. 338.

GARN, S. M.; ROHMANN, C. G.; and GUZMAN, M. A.: Malnutrition and Skeletal Development in the Pre-school Child. In: Prevention of Malnutrition in the Pre-School Child, W. Henry Sebrell, ed. Food and Nutrition Board, National Academy of Sciences, National Research Council, Washington, D.C., 1965.

GUZMÁN, M. A.; ROHMANN, C.; FLORES, M.; GARN, S. M.; and SCHRIMSHAW, N. S.: Osseous Growth of Guatemalan Children Fed a Protein-Calorie Supplement. Federation Proc., vol. 24, 1964, p. 338.

HEWITT, D.: Sib Resemblance in Bone, Muscle and Fat Measurements of the Human Calf. Ann. Hum. Genet., vol. 22, 1957–58, pp. 213–221.

MEEMA, H. E.: Cortical Bone Atrophy and Osteoporosis as a Manifestation of Aging. Am. J. Roentgenol., vol. 89, 1963, pp. 1287–1288.

MEEMA, H. E.; and MEEMA, S.: Measurable Roentgenologic Changes in Some Peripheral Bones in Senile Osteoporosis. J. Am. Geriat. Soc., vol. II, 1963, pp. 1170–1182.

NORDIN, B. E. C.: The Relation between Dietary Calcium and Osteoporosis in Different Parts of the World. A Report to the Nutrition Section of the World Health Organization. Privately printed, 1965.

ROHMANN, C. G.; GARN, S. M.; GUZMÁN, M. A.; FLORES, M.; BÉHAR, M.; and PAO, E.: Osseous Development of Guatemalan Children on Low-Protein Diets. Federation Proc., vol. 24, 1964, p. 338.

SMITH, R. W., Jr.: Osteoporotic Changes in Bone Mass with Age. Presented at the 92nd Annual Meeting of the American Public Health Assoc., Inc., New York, 1964.

SMITH, R. W., Jr.; and FRAME, B.: Concurrent Axial and Appendicular Osteoporosis: Its Relationship to Calcium Consumption. New Eng. J. Med. (in press).

SMITH, R. W., Jr.; and WALKER, R. R.: Femoral Expansion in Aging Women: Implications for Osteoporosis and Fractures. Science, vol. 145, 1964, pp. 156–157.

TROTTER, N.: A Preliminary Study of Estimation of Weight of the Skeleton. Am. J. Phys. Anthrop., vol. 12, 1954, pp. 537–552.

VIRTAMA, P., and MAHONEN, H.: Thickness of the Cortical Layer as an Estimate of Mineral Content of Human Finger Bones. Brit. J. Radiol., vol. 33, 1960, pp. 60–62.

COMMENTS

Dr. URIST. With regard to usefulness of cortical thickness measurements, I would like to know what is the biologic variability of the thickness of the cortex in ten adult females, glove size 7, 5 feet 5 inches tall, or in any other standard model?

Dr. GARN. We have completed studies involving means, standard deviations, etc., for a complete range of ages from age 1 through the 7th and 8th decades, in both sexes. With the very kind help of Dr. Trotter on Missouri Negro skeletons, we have completed such studies also on a pilot series of Japanese and Chinese. We have completed such studies also on close to 2,000 children, so far, in Guatemala We have here the means and the standard deviations for cortical thickness, we have the partial correlations, partially worked out for relationships between cortical thickness and

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what is more important, length of that individual bone rather than total size. We also have explored, since our material very nicely comes within families, the extent of genetic determination or apparent genetic determination of cortical thickness, medullary cavity thickness, and total periosteal diameter. There is some evidence also for cross-linked inheritance of cortical thickness, based on sister-sister correlations as compared to sister-brother and brother-brother, and based on father-daughter correlations compared to father-son, mother-son, and mother-daughter correlations.

Dr. URIST. I think this is the critical question, because in your control of race, you have an excellent point; this is important, as Dr. Trotter has shown in her classic study. I am asking for matched cases. Whether or not you can compare one patient with another and come up with an answer that merits this wonderful degree of precision that you have depends upon whether or not they are matched cases.

Dr. GARN. Individuals who have more bone to start with end up with more bone, even if the percentage loss is the same. Some ladies in our study start out in their late twenties at different points in the bone spectrum, and with the same percentage loss, end up at very different points in the bone spectrum. We can show you individual rate losses for women, over a 30-year period, and this is the second factor. It does appear to us that there is a point below which the structural integrity of the bone is impaired. This is the point at which the fractures occur.

How much bone the person had earlier and how much he has lost, which is then of major metabolic importance to the understanding of bone loss, can be separate from the purely mechanical considerations which suggest that after a woman has a second metacarpal cortex reduced below 2 mm, that bone, the other bones in her hand, the round bones in her hand, and the other bones in the woman are failing to continue their mechanical integrity.

Dr. URIST. Have you answered my question: What is the biologic variability? What percent would you say it was?

Dr. GARN. Women in the mid-twenties and mid-thirties (this is 153 women) have a mean of 5.4 with a standard deviation of 0.8, that is, roughly at 16% coefficient of variability. In the osteoporotic age group, for women in their 7th decade and beyond, we have a mean of 3.7 with a standard deviation of 0.7.

Dr. NORDIN. You should beware of taking a measurement like this and saying: Does this or does this not tell me if a patient has osteoporosis? This is not the way to look at this measurement. This puts into quantitative terms the amount of cortical bone present in a particular bone of the hand. There is a lot of evidence to show that this generally correlates with other bones in the skeleton, but this is really a novel issue.

In rheumatoid arthritis, for instance, cortical thickness won't correlate with the rest of the skeleton. It will just give you a measurement of the rheumatoid changes in the hand. Instead of looking at the hand and arguing as to whether or not this hand is osteoporotic, you can now look up the reference standards of Garn, Nordin, or anybody you like, and you can say this hand is outside the 95% limits of normality for this age.

Dr. URIST. The purpose of all this is to determine how much bone there is in the body. If this is a true index of the total bone mass, that is very fine. It would certainly be an easy way to determine it, but if there is a wide biologic variation from one patient to another that should be taken into consideration.

Dr. NORDIN. Nobody has yet been able to correlate cortical thickness with total bone mass, except perhaps in a few skeletons, but, in general, the variability between different bones is substantial. Broadly speaking, bone mass diminishes with age, particularly in women, and this bone diminishes with the rest, but there are a tremendous number of exceptions to this.

In some people, the femoral cortex goes before the hand, in others, the hand goes before the femur. In most, the spine goes before either of them, and you have got to accept the measurement with the limitations that it possesses.

Dr. GARN. Dr. Trotter pioneered in the studies relating the bones of the skeleton to each other and determining in skeletal material which bone was most predictive. One can extrapolate from her data to some extent, and since then she has compared cortical thicknesses of the femur to other parameters. Our group has compared the cortex of the second metacarpal to that of other bones of the hand, arm, leg, and foot. Dr. Nordin and Dr. Smith, in turn, have compared various approaches to simple cortical thickness of the hand to bone loss in the central skeletal mass.

We are well along in deciding whether we have approaches that can be clinically useful, useful in population surveys, and useful—and I think this is the most important thing—in measuring how fast bone is being withdrawn from a particular individual, and what the nutritional, metabolic, and activity correlates of these are.

Dr. NORDIN. This is absolutely true, but if you look at Dr. Trotter's interesting and excellent work, the actual predicted value of the femoral cortical area carries an error of ±30%. When you consider that we are particularly interested in the loss of trabecular bone (which constitutes perhaps something like 5 or 10% of the skeleton) and when you think that we are interested in situations in which you may have lost perhaps 20% of your trabecular bone (which is perhaps 2% of your skeleton), then an estimate of cortical mass which carries a predictive precision of ±30% is quite meaningless in terms of trabecular bone. I don't want to decry the measurement. I was one of the first to introduce it, but I want to emphasize its limitations. Its limitations are that you are measuring the metacarpal of the hand, and it so happens that

then it broadly correlates with other bones. You must not take it beyond that, for it certainly is not sufficiently sensitive to replace some measurement of trabecular bone.

Dr. WHEDON. I want to re-emphasize generally the statement that Dr. Nordin has just made. I think there is such a difference in the function of the two major classes of bone, trabecular versus cortical, that to attempt to go a long way with precise measurements of cortical bone is to be riding on the wrong horse, as far as determining what happens to changes in calcium stores in a skeleton as they appear in the increasingly common disorder, osteoporosis.

Dr. TROTTER. I agree with all of you. I think there are these variations from skeleton to skeleton, but in a series of aged skeletons, I found significant correlations in loss of bone among ten series of sets of bone from each skeleton. The skull was omitted because it was too irregular, and the hip bone was omitted. But for the long limb bones, the vertebral column in segments (cervical, thoracic, lumbar, and sacral), there was a correlation.

The Interrelationship Between Bone Density and Cortical Thickness in the Second Metacarpal as a Function of Age

NAPOLEON WOLANSKI with the collaboration of JAMES EAGEN
The Department of Growth and Genetics, Fels Research Institute

This paper reports on an attempt by the author to investigate the mineral content of bone cortex in relation to its thickness. We assumed that this content changes with age and varies among individuals of the same age.

Mineral content, which is important for the study of nutrition and of metabolic diseases such as osteoporosis and osteomalacia, has so far been measured *in vivo* in terms of bone density or of cortical thickness of a given bone (Garn et al., 1960; Garn, 1962). Cortical thickness probably correlates with the volume of the bone, which does not seem to be equivalent to the quantity of minerals inside the bone, either histologically or biochemically.

Bone density is doubly inaccurate. First of all, what was sometimes measured was not bone density alone but rather the total density of bone and soft tissue. This is of dubious value, since soft tissue may vary in thickness at the point which is being measured as well as in the percentage proportions of marrow, water, and other materials. The recent paper of Baker, Mack, Shraer, Vose, et al., involves correction for soft tissue.

Secondly, even if one considered bone density alone without soft tissue, the aluminum wedges or steps used as equivalents for the measure might vary in the composition of the metal, which might not be sufficiently pure chemically.

For these reasons, we developed a new method for the investigation of the problems. This method is similar to Mainland's (1963), with which we became familiar only after our research was finished.

METHOD

X-ray pictures of the left hand were taken with the machine at Fels Research Institute on Kodak nonscreen film made especially for the investigation of bone density. The second metacarpal of the left hand was analyzed.

Cortical thickness was measured with an especially modified pin-point caliper. First we measured bone thickness at the thinnest point of the shaft of the second metacarpal and then the width of the medulla at the same point. Cortical thickness is the difference between these two dimensions. While this measure actually represents double the cortical thickness, it is well suited for comparison with bone density. Our measurements were exact to 0.01 mm.

The thinnest part of the second metacarpal was chosen for measuring both cortical thickness and bone density, because this part is comparable in different individuals, particularly from the point of view of structure, whereas the center of the bone is not. Density was measured at midpoint between the lateral walls of the bone.

Density of tissue was defined by the optical density on the film. To determine it, we used a Weston Photographic Analyzer and a scale with

a range from 0 (0 optical ray obsorption) to 3 (total absorption). It is important to note that on film the relationships will be the reverse of the real ones, that is, where X-ray absorption is, in fact, the highest, film optical density will be the lowest, and vice versa.

Optical density measured in this manner depends on the following factors:

1. The true density of tissues (or whatever other materials are used for comparison) being X-rayed;
2. X-ray exposure technique, i.e., the amount of X-ray energy used, exposure time, distance between machine and negative, etc.;
3. Film development technique (chemicals used and their temperature, the presence of air bubbles while the film is being rinsed), secondary radiation, etc.;
4. Film density itself, that is, the density of the film-base and emulsion: minimum film density, fog;
5. Foreign objects or materials present between the X-ray machine and the film (e.g., aluminum filtration, paper cover of the nonscreen film, etc.). When Kodak nonscreen film is used, their presence is minimal.

The following symbols were used as explained below:

MFD (Minimum film density—fog)—film density at a point shielded by a 20 mm lead plate, i.e., where no X-rays could reach the film. Thus, this density is dependent on the quality of the film. In this paper we used only about 4 mm lead plate.

SD (Standard density)—film density at a point shielded by 4 mm plate of aluminum, i.e., where X-ray absorption rate would remain the same regardless of exposure techniques used. Thus film density at this point would depend on factors listed under 2, 3, and 5 above, as well as 4 (*MFD*). In this paper we used a plate of alloys of known compositions of aluminum.

TD (Total density)—film density at the point examined. This would be expressed as TD_{ST} when measured below the soft tissue, and as *TD* below both the soft tissue and the bone.

MFD, *SD*, *TD* and TD_{ST} were all measured in our study. *TD* was measured at the point already mentioned, namely, the thinnest part, midpoint of bone thickness, and TD_{ST} was taken at the same height, midpoint between the second metacarpal and the third.

After all the measurements were collected, we attempted first of all to eliminate the film fog factor. This was done by subtracting *MFD* from *TD* ($TD-MFD$), and from TD_{ST} ($TD_{ST}-MFD$).

Since our goal was to find bone density, we applied the following formula:

$$CBD \text{ (corrected bone density)} = (TD-MFD)-k(TD_{ST}-MFD),$$

where $k=$

$$\frac{\text{soft tissue thickness at the point examined}}{\text{total hand thickness at the point examined}}.$$

In the given case, total thickness at the point examined is the distance between points on the palm and the back of the hand above and below the measured point on the second metacarpal. The value of k may vary between 0.1 and 0.9. We used the value of $k=0.75$ realizing that this value changes with age; particularly it is different in children under 5. This is a better calculation of "k" for each subject (measurement of hand thickness using anthropological caliper).

The above procedure eliminated to some extent the influence of soft tissue (skin, subcutaneous fat tissue, muscle, ligaments, marrow, etc.). This elimination was not complete, since, thickness being equal, even soft tissue varies in density as, for example, in the proportion of subcutaneous fat tissue to muscle, etc. Still, it was a step forward.

Figure 1 shows changes in bone density thus corrected (*CBD*), by age. But there were other influences at work here as well, namely, those in no way related to hand tissues but rather to the techniques used in developing the X-ray film. In order to eliminate those, we used the measure SD (standard density) minus film density ($SD-MFD$).

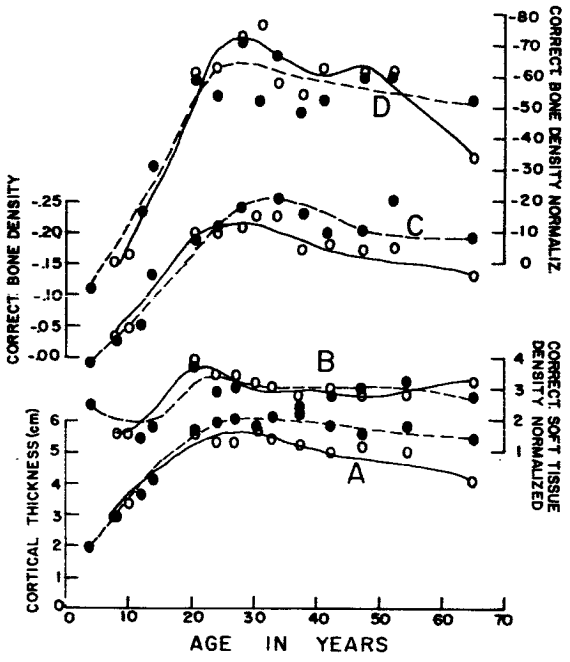

FIGURE 1.—Changes by age in second metacarpal cortical thickness (A), corrected normalized soft tissue density (B), corrected bone density (C), and corrected normalized bone density (D) in men and women at the ages of 2 to 74 years. The points represent arithmetical means; the curves are smoothed.

By dividing the results obtained by SD−MFD we get values independent of technique as follows:

a. Corrected soft tissue, normalized:

$$CSTD-N=\frac{TD_{ST}-MFD}{SD-MFD}$$

b. Corrected bone density, normalized:

$$CBD-N=100\,\frac{CBD}{SD-MFD}$$

When the above method is used, all results can be compared with one another, *on one condition*, namely, that a 4-mm aluminum plate is used for finding *SD*. One could, however, use chemically pure metal, an easily obtainable metal sample such as silver or gold (or chemically pure aluminum), conforming to some international agreement. One would then need thinner plate because of the greater atomic weight of these metals, and therefore, higher absorption.

Figures 1B and 1D show the values of *CSTD−N* and *CBD−N*.

DISCUSSION OF RESULTS

We took the above measurements from 176 male and 153 female subjects, ranging in age from 2 to 74 years. All subjects were permanent residents of southern Ohio (region near Yellow Springs).

Figure 1 shows the smoothed curves formed by the arithmetical means of the values under discussion, namely, cortical thickness (*CT*), soft tissue density between the second and third metarcarpal (*CSTD−N*), and bone density at the point described, on the second metacarpal (*CBD* and *CBD−N*).

As we see from figure 1A, cortical thickness increases at a particularly fast rate between 3 and 20 years of age. Bones are thickest at about 24 years of age in women and 28 in men. After that we observe a decrease in cortical thickness (bone loss). From puberty on, bone cortex in boys is thicker than in girls. This may be related to the boys' taller stature, hence larger bones. We did not calculate cortical thickness in relation to bone length.

Soft tissue density (*CSTD−N*), after some decrease at under 10 years of age, increases rapidly until the ages of 20 to 23 (fig. 1B). Then it again decreases rather quickly at about 28 years of age in men and about 35 in women. After that it remains at a more or less steady level. Women between 15 and 27 years old have higher soft tissue density than men; later in life the reverse is true.

Unadjusted bone density (*CBD*) varies greatly from the corrected, particularly as the two sexes (*CBD−N*) are compared. This can be understood easily if we keep in mind that in X-ray examination X-ray dosages vary depending on hand thickness. These differences are adjusted through the *SD−MFD* technique.

Corrected bone density, *CBD−N* (fig. 1D), increases rapidly between the ages of 3 and 22 years, and more slowly up to about 30 years of age. After that it begins to decrease slowly but

steadily in men, and slightly more rapidly in women, particurlaly after the age of 50, i.e., in the postmenopausal period. Up to approximately 20 years of age, bone density is higher in boys than in girls, between 20 and 55 it is greater in women than in men, and vice versa once more, above that age.

THE INTERRELATION OF BONE DENSITY TO CORTICAL THICKNESS

In order to evaluate bone density with relation to cortical thickness, we used an index of corrected bone density (index *CBD*), and a normalized index *CBD−N*, obtained through the following formulae:

$$\text{Index } CBD = 10\frac{CBD}{CT}$$

$$\text{Index } CBD-N = \frac{CBD-N}{CT}$$

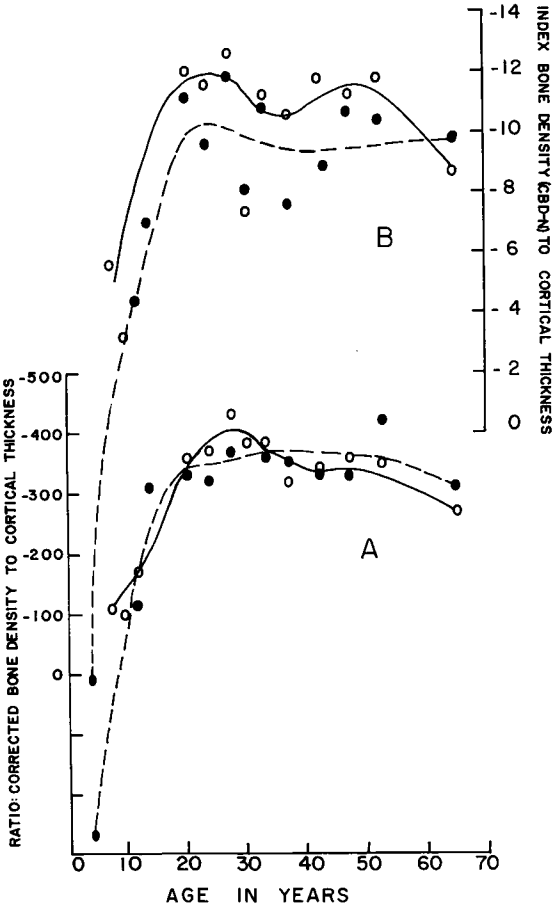

FIGURE 2.—Changes by age in the relative bone density of the second metacarpal, expressed as the index of corrected bone density (A), and the index of corrected bone density normalized (B), in men and women at the ages of 2 to 74 years. The points represent arithmetical means; development curves are smoothed.

Clear differences are descernible between *CBD* and *CBD−N* (fig. 2). We shall use index *CBD−N* for analysis.

As we can see (fig. 2B) up to 25 years of age bone density in subjects of either sex increases rapidly with respect to cortical thickness. It decreases between the ages of 25 and 37. Further increase in density can be observed between the ages of approximately 37 to 50 in men, for whom it then remains on a steady level, and between about 37 and 48 years of age in women. In women from about 50 on, relative bone density decreases rather rapidly. To compare between the sexes, the changes are similar for both up until about 50 years of age, although relative density, as expressed by the *CBD−N* index, is consistently higher in men. From 57 years on, during the postmenopausal years in women, their relative bone density becomes lower than that of men.

This phenomenon shows up with particular clarity on a scattergram of correlations of cortical thickness to bone density (*CBD-N*), as illustrated in figure 3.

We see a fairly regular rate of growth of cortical thickness and bone density with age, from 2 to about 27–34 years in men, and between 28 and 31 years in women. The parallel lines indicate greater thickness in men than in women of the same age but greater density in women.

Of special interest, however, are changes occurring after maximum density (*CBD-N*) has been achieved. These are different for each sex. In women, the process consists of a rather sharp decline in bone density and a slow decline in cortical thickness. Thus, while differences exist in rates of change, their direction is generally the same. The process runs a slightly different course in men. Their cortical thickness remains

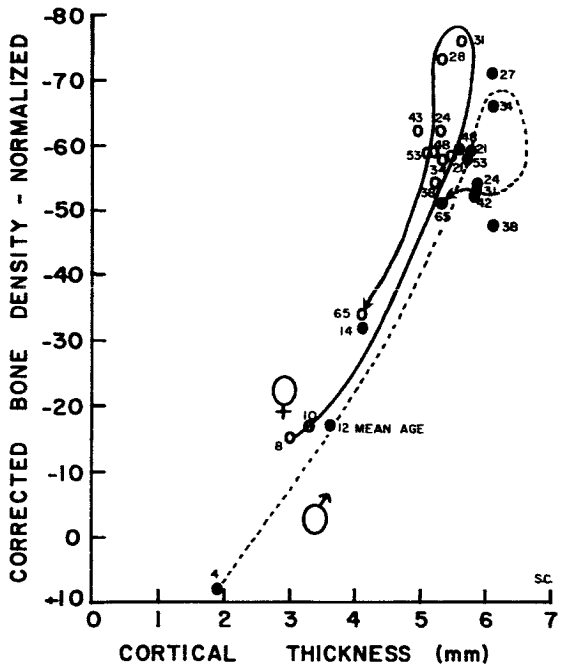

FIGURE 3.—A scattergram of the cortical thickness (*CT*) and bone density (*CBD–N*) of the second metacarpal. Points represent arithmetical means for given age groups. The course of the curve is semi-schematic.

the same even after bone density has begun to decrease. This happens between about 34 to 38 years of age, after which the decrease in cortical thickness is accelerated while bone density remains on a steady level.

CONCLUSIONS

It appears that the method presented allows for a more exact examination of the process of bone mineralization *in vivo*. Of particular significance may be the analysis of cortical thickness (*CT*) and bone density (*CBD–N*) from a scattergram. The method appears to be particularly apt for the diagnosis of cases of osteoporosis as opposed to osteomalacia, as well as other diseases of bone metabolism, e.g., *fragilitis ossium*, *osteitis fibrosa cystica generalisata*. It may be particularly useful in distinguishing between cases of disturbances of protein metabolism and those of calcium-phosphorus metabolism.

Many aspects remain unexplained, such as the different course of changes in men and women after the ages of 27 to 30 (fig. 3), and particularly the differences in direction of change in bone density (*CBD–N*, fig. 1D) in the postmenopausal period, that is, when there is a lack of estrogen.

For explanation of this phenomenon, one would have to resort to a histochemical and microanatomical examination of bone tissue. There may be differences in changes that are microstructural in nature, as those in the chemical composition of bone tissue.

It would also be of interest to evaluate the evidence that there are two periods of particularly high bone density (index *CBD–N*), the one occurring at about 25 years of age—which is probably the period when progressive bone development comes to an end—and the other at about 48 years of age in women and about 55 in men, which may be the time when intensive bone calcification comes to an end.

ACKNOWLEDGMENTS

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REFERENCES

GARN, S. M.: An Annotated Bibliography on Bone Densitometry. Am. J. Clin. Nutrition, vol. 10, 1962, pp. 59–67.

GARN, S. M.; HARRISON, H. E.; and MASON, K. E. (eds.): Transcript of the Workshop on Bone Densitometry Held at the Stone House, NIH, December 4–5, 1959, Yellow Springs, Ohio, 1960.

MAINLAND, D.: X-Ray Bone Density of Infants in a Prenatal Nutrition Study, The Milkbank Memorial Fund Quarterly, vol. 41, no. 4, 1963, pp. 1–106.

COMMENTS

Dr. NORDIN. Dr. Wolanski has made a very good fundamental point which I tried to raise this morning with Dr. Schraer, that is that the difference between bone densitometry and cortical thickness was not brought out this morning. One would want to know what densitometry tells one after correction of the bone for cortical thickness. There are two different things here. The amount of cortex in millimeters, and the quality or nature or density or whatever it is, porosity, within that cortex. I think I am right in saying that Dr. Wolanski has brought out this particular point, which I think is of very considerable interest and which I think has not been touched on.

Dr. GARN. I think that many of us are interested now in whether or not the aluminum density equivalent per millimeter of cortical bone thickness changes. We have examined this in part again with Mildred Trotter's material. The question now is this: Is the material we measure and report as cortex the same material from age 20 to age 70?

Dr. ARNOLD. Would an increasing porosity in females to a greater extent than in males explain this? In other words, would this give you a decreasing density when normalized to a cortical thickness?

Dr. NORDIN. No, it is the other way around, as I understand it. That is why I wanted Dr. Garn to explain it to us, because it looks to me the opposite from what we would expect. As I understand it, if Dr. Wolanski has done what he wanted to do, it is exactly the opposite from what we would expect.

Dr. GARN. I think, Dr. Nordin, that we are both beginning to agree that men are losing bone as well as women, and I think that, to some extent, the rate at which men lose bone has been underestimated because of the attention given to women and their more severe complaints.

Dr. ARNOLD. That has been the pathological position from the beginning, that men atrophy at about the same rate as women.

Dr. GARN. You see, if you would take one of these estimates, which none of us quite like to make, and extrapolate from the little bit of bone that we have all started on (one little bone in the left hand), this extrapolation would indicate that the amount of bone lost is as great in the male as in the female, whereas, of course, we all know that the percentage loss is greater in the female than in the male.

Dr. NORDIN. But that does not explain this graph, which suggests that the female has more density for cortical thickness than the male.

Dr. GARN. This is something that Dr. Schraer reported years ago for the middle section of the middle segment of the fifth digit, which has been ignored. I thought he was totally wrong. It did not make sense. But when one considers the total area, women with smaller tubular bones have, for those smaller tubular bones, more cortical material in relationship to the bone area than men do.

Dr. NORDIN. In other words, the cortical material itself is less porous in these women than in men. This is a very remarkable thing. This is a paradox. That is why I was hoping a language problem might have occurred here and it might have been interpretable.

Dr. SMITH. Just to add to this point, we have made observations on density of rib, subperiosteal compact bone, iliac compact bone, and femoral compact bone and compared them between sections; that is, by taking cylinders of known length and known diameter and weighing them to fractions of milligrams. We cannot find significant differences in actual density of compact bone in respect to this question between the sexes, when age matched.

Dr. URIST. Assuming that this observation is true, microradiography of cross sections of the cortical bone might show more interstitial bone of greater density in the female. There is more calcium, there are calcified osteocytes and plugged up blood vessels, and this has been interpreted as a manifestation of osteoporosis as a disease. Whether it is or not is another question. I think we are still on the subject, really, of physiologic osteoporosis. I don't think we have gotten into the subject of the clinical problem, but the answer might be in a study of the microanatomy.

Dr. WHEDON. Dr. Smith, do you want to say anything with regard to the findings you have made with respect to the expanding cortical diameter with age?

Dr. SMITH. We have not found expanding cortical diameter with age in the second metacarpal, as Dr. Garn has indicated, measuring in essentially the same area, but we do find it in the mid-shaft of the femur. I think this is one of the blessings of having a simple millimeter scale that you can hold up to the X-ray, and take direct measurements. You can uncover this very remarkable gain of about 3.5 to 3.6 mm of external diameter gain at mid-shaft femur, thus your presentation this morning of providing a ratio of cortical bone to diameter, I think, may not be wholly valid for those bones wherein this phenomenon is taking place. In the metacarpal I think this is true, and I am just waiting for Dr. Garn or someone else to confirm this very interesting phenomenon, which is quite substantial, in the mid-shaft of the femur.

OTHER METHODS OF DENSITOMETRY

CONSTANTINE J. MALETSKOS, *Chairman*

I am going to make my remarks quite short in the interest of proceeding with today's session.

Yesterday we talked about X-ray densitometry which involved the physical measurement of transmission of electromagnetic radiation through matter. We have a variety of different kinds of electromagnetic radiation, and today's session is going to examine some different aspects. Again, it will be fundamentally a transmission measurement, but since there will be a different situation, there will be different problems arising. Perhaps there will be advantages and perhaps there may be disadvantages. It might be possible, for example, to make more direct corrections for tissue absorption than could be made by the method that was discussed yesterday.

The session today also includes the use of other physical agents which allow you to make such a measurement, and one of the later papers will involve the technique of ultrasound. I think it is clear from yesterday's discussion that there are a number of variables involved in the final assessment of this situation, namely, what has happened to bone under different circumstances. We have to consider bone size, bone density, bone mass, and even bone composition, and we have to consider this both from the points of view of the mineral and of the organic matrix.

As we have our talks and discussions, I think it would be well worthwhile bearing in mind that each person should try to be clear as to what he is referring. It unfortunately seems to me that we will have to have some information on every aspect to really make a final assessment. The situation is more complicated than we would like it to be, but this is part of life.

We have other problems that are involved with respect to precision and accuracy. From a strictly research point of view, we would perhaps like to have this situation be as accurate as possible. From a clinical point of view, we perhaps might not need this high degree of accuracy and precision, and we should bear this in mind in our discussions. The clinician wants to treat a sick person and make him well. The researcher wants to find out more about the mechanisms. We want to be careful that we do not apply the precision and accuracy to a situation of clinical use that would normally be applicable only to a research situation.

I would like to conduct the meeting as we did yesterday. I think this resulted in a lot of fun where there was much free discussion between the speakers and the participants. Later on in the afternoon I would like to bring up the subject of linearization again to see if we can resolve that to almost everybody's satisfaction. It would be a great accomplishment if we could leave this meeting with everybody happy in this respect.

Finally, since Dr. Jenkins has invited other thoughts in the measurement of bone density, I would like to take a couple of minutes to tell you how the technique of neutron evaluation can be used in this field and to let you know that this technique does exist and is now in its infancy as to development.

Perhaps measurements of bone density could be made while man is in flight. It might be worthwhile thinking about this in the type of techniques that we have, because this would be the ideal situation.

I personally think that neutron activation analysis might be the technique, but there are other ways of doing it.

I would like to start now with the morning session. We are going to be discussing the technique of the use of isotopes. The first three speakers are going to be talking more or less on a similar subject, looking at it from different points of view.

C. J. M.

Bone Mineral Measurement by Improved Photon Absorption Technique

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Departments of Radiology and Physics,
University of Wisconsin

The direct photon absorption technique for measuring bone mineral content (Cameron and Sorenson, 1963) has been tested for reproducibility of results and for reliability as an indicator of actual mineral content. An improved bone-scanning system has been constructed for *in vivo* measurements on humans, and a series of experiments were performed to observe the effects of various instrumental factors on the measured mineral content.

Mazess et al. (1964) used the technique on the tissue-covered bones of cadavers and obtained a correlation coefficient of 0.96 between measured bone mineral content and subsequently determined bone weights and ash weights. He concludes that the technique is the most accurate yet demonstrated for determining bone mineral content in the presence of overlying tissues. The technique was also used in a study of dietary-induced changes in bone mineral content in chickens, in the University of Wisconsin Poultry Science Department (Mantilla et al., unpublished observations). In these studies the correlation coefficient between the measured mineral content and subsequently determined ash weights was 0.95 and 0.99 for measurements made *in vivo* and *in vitro*, respectively. All of these studies were done with the earlier scanning apparatus.

The improved scanning system was used for repeated measurements of the bone mineral content of the left radius of a normal 26-year-old male over a period of several weeks. The results are shown in figure 1. Each point represents the average of three to six consecutive measurements. The error bars indicate the standard deviation of each set. The 1.3% standard deviation of these averages indicates the reproducibility of this system.

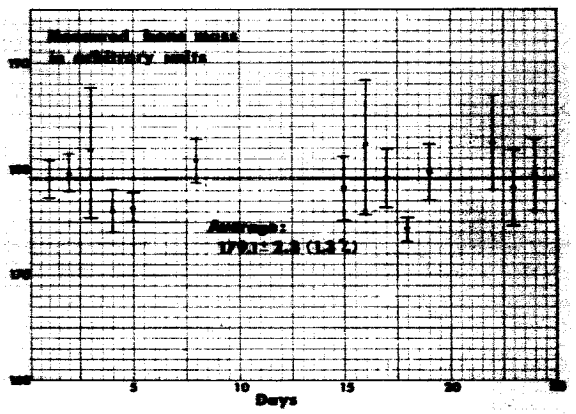

FIGURE 1.—Results of repeated bone mineral determinations on a normal male. Each point is the average of 3 to 6 measurements.

The improved system can be used for *in vivo* scanning of such human bones as the radius, the metacarpals, and the phalanges (fig. 2). The device can also be oriented to scan in a vertical plane and can be adapted for other bones, such as the femur, os calcis, etc. The radioactive photon source and detector are mounted on a turntable at a radius of about 9 cm. The scanning motion is obtained by driving the turntable on its circumference with a slow speed synchronous motor through a rack and gear system (fig. 3). The scan path is curved rather than linear; but the increase in the apparent

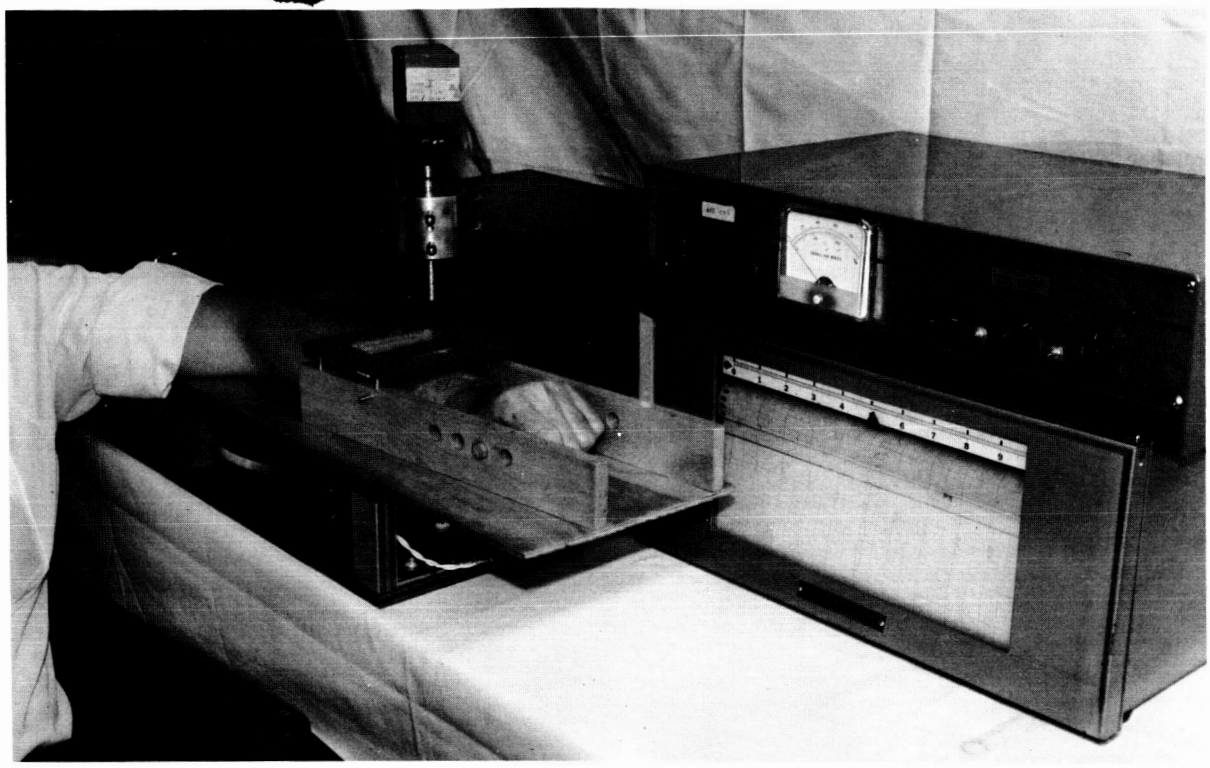

FIGURE 2.—Improved scanning apparatus in operation.

FIGURE 3.—Photon source and detector on turntable.

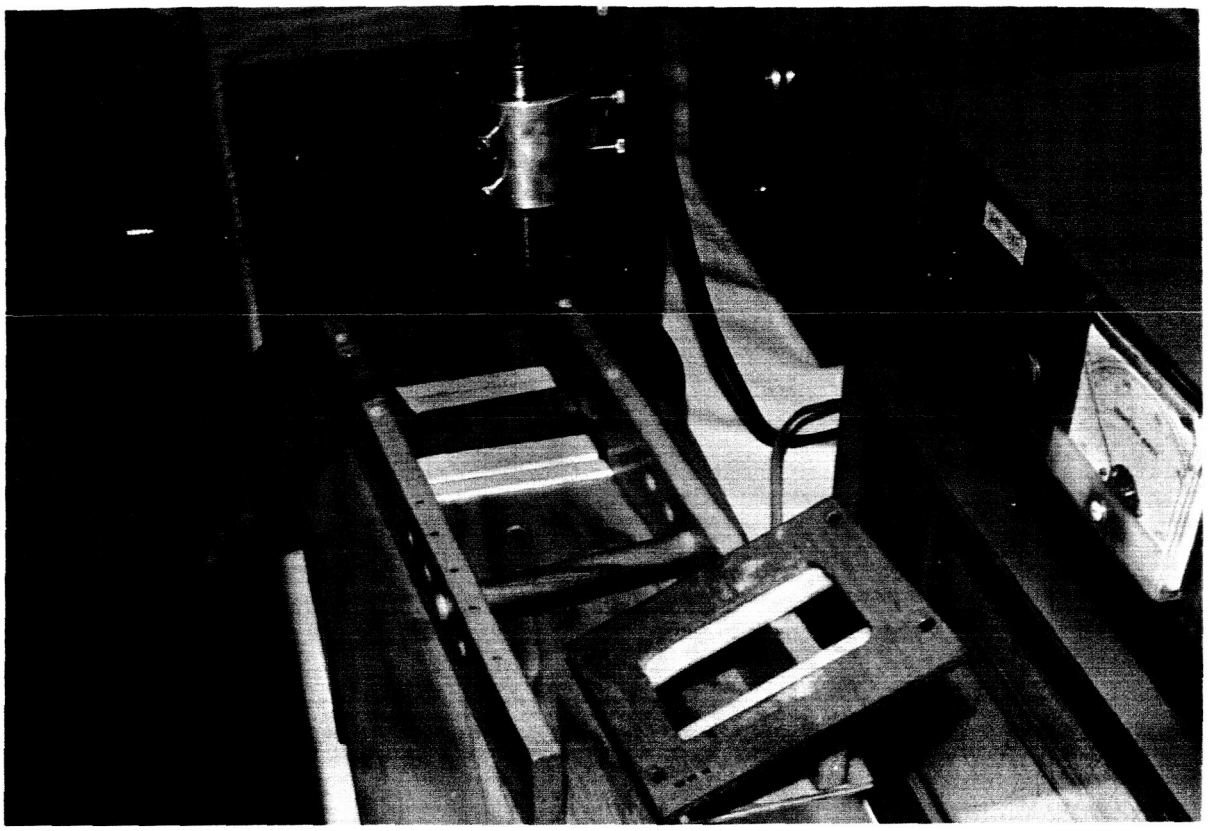

FIGURE 4.—Scanning apparatus showing rubber tubing, compressor plate, and arm holder.

width of the bone is less than 1% for diameters up to 2 cm.

To scan the radius, the forearm is placed in a wood and plexiglass holder. A rigid board running the full length of the forearm and a hand-grip, which can be placed at various distances from the scan path, are used to obtain reproducible positioning (fig. 4). The part of the arm to be scanned is encircled with a section of 7/8-inch diameter flexible rubber tubing (Penrose drain), filled with a 2% solution of potassium sulfate. The tubing is then compressed between parallel pieces of plexiglass to provide a constant reproducible thickness of tissue, tissue equivalent, and bone throughout the scan. The compressed rubber tubing also helps to keep the arm stationary. Using the 2% potassium sulfate solution, we obtained uniform absorption on both sides of the bone. Linear scanning speeds of 0.54, 0.79, and 1.58 mm/sec are presently available by changing drive gears. Typical scanning times are about 30 seconds, using the 0.79 mm/sec speed.

A 35-mc I^{125} photon source (27.3 keV) was constructed for use with this system. Twelve beads of Dowex 1 x 4 ion-exchange resin (20–50 mesh) were placed in a 1-ml solution of carrier free radioactive iodine of 50 mc activity for about 4 hours. The beads were periodically removed from the solution to follow their increase in activity. The beads did not reach saturation during the 4 hours as indicated in figure 5. A more active source probably could have been made in the same time by starting with a solution of greater activity.

The radioactive resin beads were placed in a 1-mm diameter x 12-mm deep hole in a brass holder. The beads filled the hole to a depth of about 7 mm. The photon beam is thus collimated by a 1-mm diameter x 5-mm deep aperture. A single layer of 0.06-mm tin foil provides beam softening filtration. Tin has its k-absorption edge at 29 keV and it preferentially absorbs the 31 and 35 keV components of I^{125}.

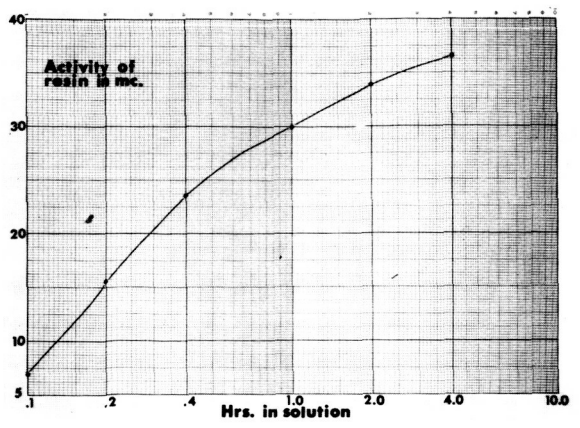

FIGURE 5.—Activity of resin beads in radioactive iodine solution.

The source is viewed end-on by the detector and has an effective cross section of 0.7 mm².

The NaI(T1) scintillation detector, 1-cm diameter x 2-mm thick, is mounted on a 1-inch photomultiplier tube. The output of the photomultiplier tube is sent to a pulse height analyzer and then to a ratemeter with a linearity of better than 1%. The output of the ratemeter is transmitted to a potentiometric recorder having 0.25% accuracy. Photon detection rates are read directly from the chart record. We use a ratemeter time constant of 0.3 second, with a linear scanning speed of 0.79 mm/sec. When the time constant is increased to 1.0 second, the measured bone mineral decreases by about 2%. Decreasing the time constant to 0.1 second does not have a significant effect on the measurements.

We plan to replace the ratemeter and chart recorder in our present system with a digital ratemeter and high speed printer in the near future. This will eliminate problems arising from the use of different ratemeter time constants and errors in reading the chart records. We expect our results to improve with this change. The digital ratemeter will also be adapted for use with an electronic computer for rapid evaluation of our data.

We have performed a number of experiments to examine the effects of the soft tissues surrounding the bone on the measured mineral content. Glass slabs and test tubes were used to simulate bone, and slabs of lucite to simulate soft tissues. The 35 mc of I^{125} described above was used as a photon source. The detector was collimated with a 1/8-inch diameter x 1-inch long aperture in lead. The source to detector distance was about 6 inches, which is typical for measurements on the radius.

With an analyzer base level of 25.3 keV and a window width of 4 keV, we measured the "bone mineral content" of a glass test tube (1/2-inch diameter with 1/32-inch wall thickness) for various thicknesses of overlying lucite. The 1/2-inch lucite slabs were placed between the test tube and the photon detector. The results are shown in figure 6. The measured "bone mineral content" varies by about 10% under these conditions.

We measured the absorption (I_0/I) of the 27.3 keV photon beam in a 7/32-inch glass slab, for various analyzer base level and window width

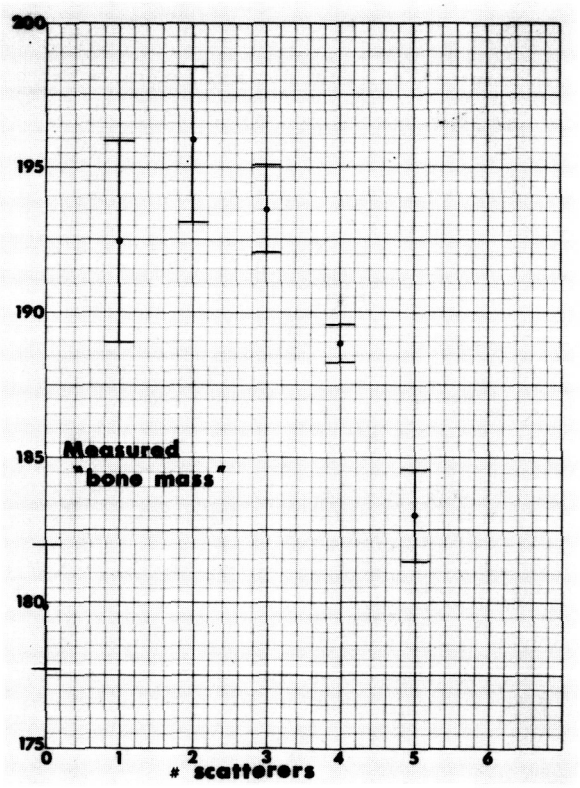

FIGURE 6.—Effect of lucite scatterers on measured "bone mass." Each scatterer is 1/2 inch thick.

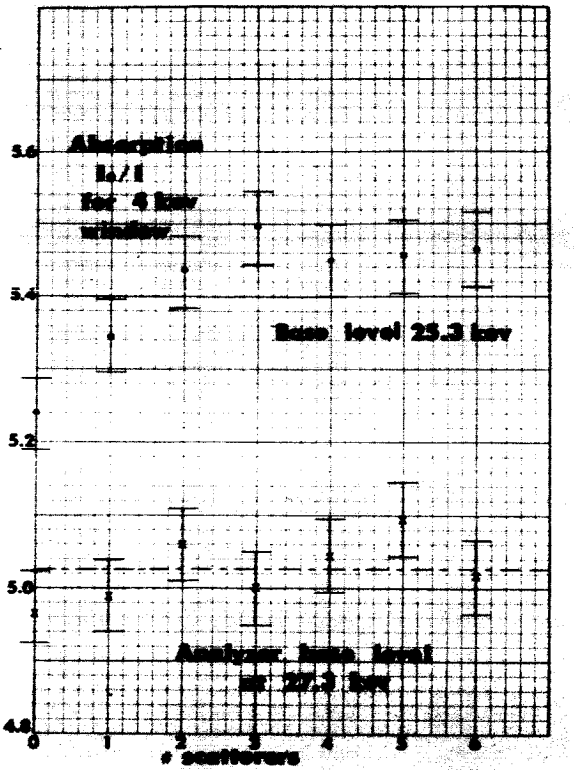

FIGURE 7.—Effect of lucite scatterers on measured absorption for two analyzer base level settings. I^{125} (27.3 keV) photon source.

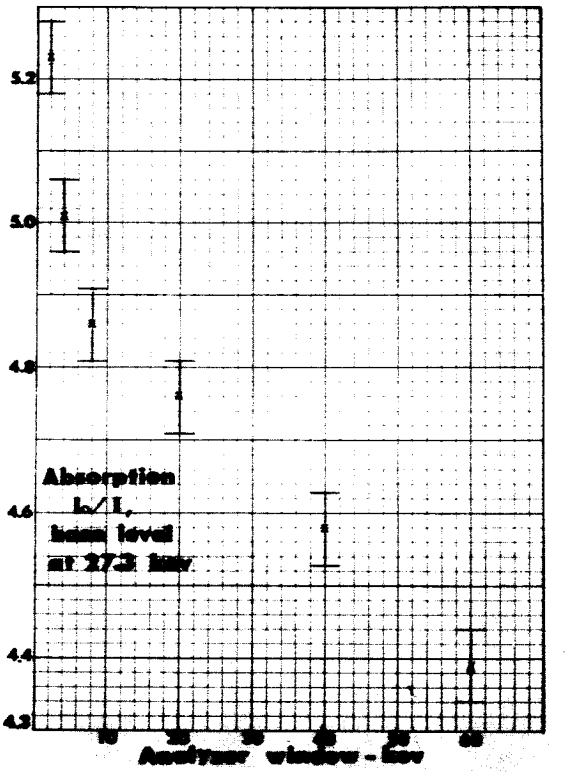

FIGURE 8.—Effect of window width setting on measured absorption, with a constant number of lucite scatterers (2 inch total thickness).

settings, in the presence of different thicknesses of overlying lucite scatterer. Figure 7 shows the absorption vs. number of ½-inch lucite thicknesses for two analyzer base levels. The absorption measurements were constant within statistical limits when the analyzer base level was set at 27.3 keV.

Figure 8 shows the apparent absorption in glass for different window widths, and a constant analyzer base level of 27.3 keV. A 2-inch thickness of lucite was present in these measurements.

The experiments illustrated by figures 5–7 were repeated for other relative positions of the lucite scatterers, e.g., for the lucite scatterers placed between the source and the absorber. The results were similar to, but not the same as, those obtained for the original positions. This rules out simple correction factors based only on the knowledge of total thickness of the scattering medium, since the relative position of the absorber in the medium is also important.

The results of these experiments indicate that for accurate measurements of bone mineral content by the photon absorption technique it is necessary to use a pulse height analyzer, with carefully chosen operating conditions. For best results the analyzer base level should be about 27.3 keV, and the window width only a few keV. It is also important that these settings be the same to obtain reproducible results. (This work was supported in part by the USAEC, under contract number AT-(11-1)-1422.)

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REFERENCES

CAMERON, J. R.; and SORENSON, J.: Science, vol. 442, 1963, p. 230.

J. MANTILLA, S. BABCOCK ET AL. To be published.

MAZESS, R. B.; CAMERON, J. R.; O'CONNOR, and KNUTSEN, D.: Science, vol. 145, 1964, p. 388.

COMMENTS

Dr. MALETSKOS. Your detector does not move now?

Dr. CAMERON. Yes, the detector and source are on a turntable. They move around an arc. You might say that is not a straight line, but the error there is less than 1% for a diameter of up to 2 cm or so. It is an extremely small error and well within the accuracy with which we are trying to work.

Dr. MALETSKOS. You chose the turntable because you could get more drive that way?

Dr. CAMERON. It was chosen for simplicity. This screw drive, this split nut is a nuisance. There is some give between them. They are not as reproducible as a good rack and gear system. This system also is easy for changing speeds. All we have on this is a timer motor, all three different gears. We can put other timer motors on other gears around the periphery of this turntable. Thus we can have a dozen different speeds very easily.

Dr. MALETSKOS. How do you account for the fact that soft tissue is variable in terms of the effect you get here, because you have fat tissue and muscle tissue involved. Even though you are trying to get the same thickness with your liquid device, the composition of the tissue is changing from person to person.

Dr. CAMERON. That is right. This is a limitation of the technique. No technique is perfect. There are problems and there are ways in which these things can be improved. Some of the data here is less than 1 month old. I think all of our recent data are new to everybody here. I want to emphasize again that we are working a different magnitude and with a completely different system than we were with film. If I were to show this in a more optimistic method, I would have left a zero there and showed practically no change. You have to exaggerate it in order to see that there is a physical effect.

Dr. KLAPPER. Which anatomical site did you use with your technique?

Dr. CAMERON. We can use either the right or left arm. So far we have not gone beyond that. With this faster scan, we have some animal experiments using the phalanx and the second metacarpal as well as the radius and the humerus. We can do anything in this general area and we can move down to the leg. We can take the equipment and rotate it so that it rotates across the thigh. For that research, we would probably use Americium 241 because there is higher energy for the 60 keV photon.

Dr. KLAPPER. How do you achieve reproducibility since the technique, as is also true of the comparable approach of Williams and Odlum, is essentially blind for a relatively large long bone? You would be able to reproduce positioning within a centimeter or so, but for a small bone such as the middle segment of the fifth digit or for the second metacarpal, the positioning error is more of a problem.

Dr. CAMERON. I would like to relay the answer to that question to the Chicago group since they have a very good system. We have actually isolated it for positioning of the fingers well within a millimeter for their measurements on the metacarpus. We do not throw the X-ray sets away! We use X-ray films to get positions. We have not done this on our present equipment, but we will be able to X-ray and get within a fraction of the millimeter where we have taken our record on an X-ray film. The source, which has the same energy in Japan, Australia, the United States, or wherever, is an excellent simplification. We had many development problems. The detector systems are generally available. A Picker magna scatter could be modified to scan across the bone with this technique. An attachment takes actual measurements across almost any bone, across the hip bone. There are a lot of things to study on this technique. I hope as a result of this conference there will be more people working with it.

Dr. SCHRAER. I personally feel that the ultimate direction of this whole area will be as Dr. Cameron described it. I think it will be the only practical method in the future. I want to ask a few questions, however.

How much do you think equipment like this will cost to make commercially?

Dr. CAMERON. I would guess about $5,000, depending on how fancy you want to be. The only thing you need in many places is a mechanical device. Many places have the pulse height analyzer and recorders. You can get in the business cheaply if you have the Picker scanner.

Dr. GARN. There is a firm in Knoxville, I believe, that has made several complete machines for the group at the University. They are willing to make more of them. It is completely integrated with the Cracow equipment, equipment for moving either the part or the scanner uniformly built in. It is about 3

ft. long x 3 ft. wide. Their first two machines, I believe, cost about $10,000.

Dr. CAMERON. If you want to build your own, you can do it for a much smaller price.

Dr. RICH. There is one hesitation I have about using commercial radioisotope scanning equipment. We have found that the speed of the scanning device is not well controlled. In fact, the error runs around 8%.

Dr. CAMERON. Yes, you would have to watch some of these things. I am not recommending it. I am just telling you what you could do if you wanted to get started.

Dr. SCHRAER. I think if you select bones where it is not critical, you can go long distances on a femur, for example, where you won't get much change. If you avoid areas like joints, it becomes negligible. For joints it would be very important, because you have a sclerotic area and a non-sclerotic area, and if you move a half millimeter, you are in trouble.

Dr. WHEDON. What you are showing is reproducibility. This really has nothing to do with how much bone is in the arm.

Dr. CAMERON. It has something to do with it if the human will let us take a slice of his arm, if we could calibrate it, or we could take another bone and use the same thickness of scatter, a dry bone, and put in a phantom. I would guess we could hit within absolute units; we could come within 3 or 5% in absolute units of the amount of bone mineral there. The greater error in this case is due to uncertainties in composition of the bone marrow, in fat versus muscle, and some of these other factors.

Dr. SMITH. I can see how you position the arm for motion back and forth, but how do you avoid rotation?

Dr. CAMERON. If the arm is resting across a bar, there is relatively little rotation.

Dr. BABCOCK. On a human radius, it does not make a great deal of difference.

Dr. CAMERON. It would have some difference because of the size of the beam and so on. The work that Dr. Babcock will present used the older equipment. I am sure we can do somewhat better on this newer equipment. This is designed mostly for human work, but it can also be used for experimental animals.

Dr. RICH. Do you anticipate any possibility of applying this to vertebrae?

Dr. CAMERON. We have thought of using it on the head of the femur. On the vertebrae it is a problem worthwhile looking into, but I think it would be difficult. There are more interesting things to study than vertebrae, such as the thigh soft tissues.

Dr. LANZL. On Sorenson's arm are the vertical units, your centimeters, squared?

Dr. CAMERON. No, it is more complicated than that. Basically you can think of it as centimeters squared on a graph paper. He actually does this numerical integration, looks up the logs of the count, and records the numbers from a chart. He gets about 40 numbers and he looks up their logs and integrates the logs. In this way he gets a number proportional to the area on the old system. We no longer use a planimeter as such.

Dr. LANZL. In your scattering experiments, did you use glass?

Dr. CAMERON. Yes. We should ideally use something closer to bone, but we didn't expect to see that much change. Maybe now we will go back and do it with bone to see how much change there is.

Dr. LANZL. You would attribute this to a scatter component?

Dr. CAMERON. It is probably multiple scatter. Multiple scatter is something that is really difficult to work with. Single scatter is no problem at all. We have analyzed that part. When we get more than one scatter of a photon, it gets pretty difficult analytically.

Dr. LANZL. On the basis of this, would you say to avoid this problem, which is not a sizable one, would you think in terms of going to a smaller organ rather than something thick?

Dr. CAMERON. I think as long as you keep your thickness continuous; if there is an interesting study to be made of the femur, I think you should go ahead and make it. If you have to take a greater error, say of 3%, you just take it, that is all.

Dr. SCHRAER. Could you do the alveolar bone with your technique?

Dr. CAMERON. I am pretty sure you can do almost anything. We can get a protected source inside the mouth and beam it out.

Quantitation of Bone Mineral Measurement in the Domestic Hen

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Department of Medicine, University of Wisconsin, Veterans Administration Hospital, and Poultry Science Department, University of Wisconsin

Part I

The measurement of bone mineral content by the Cameron-Sorenson technique involves attenuation of a mono-energetic photon beam from I^{125} by the interposed bone and detection with a scintillation counter (Cameron and Sorenson, 1963). As with any new technique, certain questions may arise concerning the parameter that is measured. Prominent among these are questions of accuracy, reproducibility, sensitivity, components of the measurements, and the possibility that the measurement is an artifact. Mazess et al. (1964) have used this technique in human cadaver material and compared it with the weight of ashed bone segments. The correlation coefficient of these two measurements was 0.96. The present study was designed to confirm and extend analysis of this parameter by the application to an experimental animal.

METHOD

The domestic hen was selected as a test animal for several reasons: It has a long, linear bone, the tibia, which is easily accessible; it has a very labile calcium metabolism; and it will lie quietly without anesthesia for a moderate length of time. Measurements were made at a site 3 cm proximal to the tibio-tarsal joint of the left leg. The animal was placed in an adjustable holder with the left leg extended perpendicular to the scanning apparatus and the right leg held back parallel to the body. The wings were interlocked and held behind the back rest with suitable weights. By covering the hen's head with a dark cloth, it will lie quietly for periods of 30 minutes or more. The leg to be measured was held by rubber bands and modeling clay.

The measuring equipment was basically that originally described, with the following modifications: A thin tin filter was placed over the I^{125} source which removes a large percent of higher energy radiation, as just described by Cameron; the scan was continuous and read-out with a ratemeter and chart recorder rather than interrupted for scaler read-out at 1-mm intervals; and the data were calculated by summation of the logarithms of values rather than plotted on semi-log paper with planimeter measurement of the area within the scan. The leg was scanned four times without removing the animal, and the mean of these values was taken as a measurement. The standard deviation of these scans serves as a check on arithmetic errors as it seldom exceeds 3%, depending somewhat on source strength. Bone mass is expressed in operational units.

REPOSITIONING ERROR AND REPRODUCIBILITY

Measurements at various sites along the tibias of three hens are shown in figure 1. Values at corresponding sites on left and right legs are given and show fairly close agreement. The segment of bone from 3 cm to 5 cm proximal to the tibio-tarsal joint did not vary more than 5%. Hence, exact positioning for subsequent

measurements, although desirable, does not introduce large amounts of error. It might be more desirable to choose a site for routine measurement 4 cm proximal to the joint rather than 3 cm, as there is greater uniformity around the 4-cm site.

Reproducibility was measured by sequential measurements at the 3-cm site in a hen, with removal and repositioning ten times. In this instance each measurement consisted of only three scans. Figure 2 shows the ten values with their individual standard deviations. The mean and range are given in the shaded figure; the percent standard deviation for these ten measurements was 4.2%. Since an unusual degree of fatigue due to the duration of measurement contributed to motion, this value is probably higher than ordinarily encountered. Source strength and number of replications both contribute to this high figure.

ACCURACY

The accuracy of the measurement in a living system was evaluated by making 18 measurements at various sites along both left and right tibias of four hens *in vivo*. Measurements were subsequently made after the animals were killed and also after the bones were dissected free from flesh. Segments of bone were cut, using a special holder with paralleled edges. The segments were accurately measured with a micrometer, dired at 100°C to constant weight, weighed, defatted with ethanol-ether extraction in the Soxhlet apparatus, weighed, ashed for 24 hours at 600°C, and weighed. Figure 3 gives the relation between weight of 1-cm ashed segments and bone mass measured *in vivo*. The correlation coefficient was 0.957 and the regression equation, as determined by the least squares method, indicates that the operational bone mass unit used in this laboratory is equivalent to 11.5 mg of bone ash per linear centimeter of bone. Comparison of bone mass measured in bones dissected free of flesh with weight of segments gave a correlation coefficient of 0.995. Correlation coefficients between measurements made on dead animals with flesh intact were of the same order, as were comparisons with the fat-free weight of bone segments.

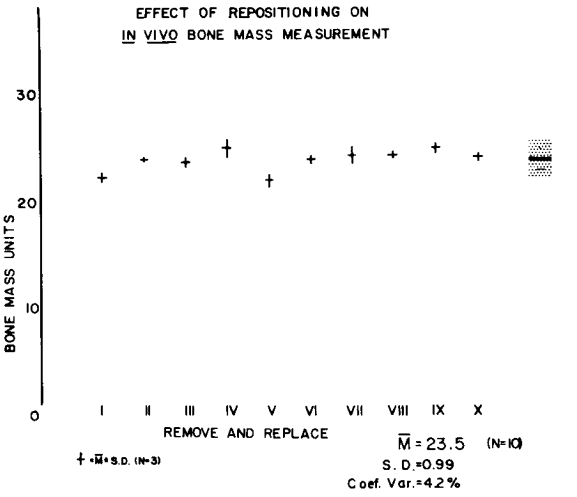

FIGURE 2.—Error of repositioning: ten measurements of same hen.

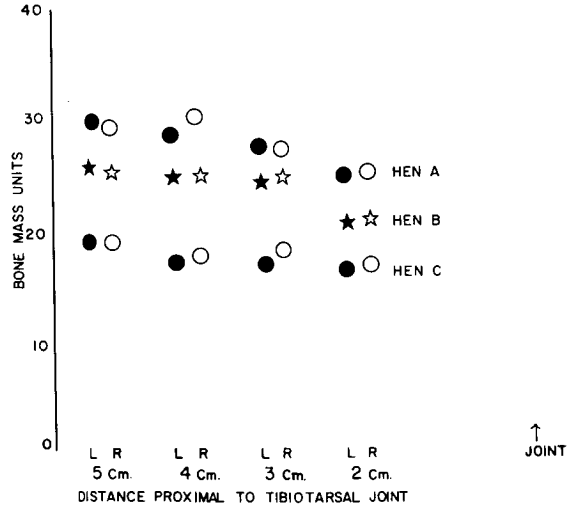

FIGURE 1.—Bone mass measurements at various sites along tibiar sites of three hens, with comparisons between left and right legs.

COMPONENTS OF THE MEASUREMENT

Measurements of bone mass on segments of human cadaver bone after defatting and ashing indicate 89% of the bone mass measurement represents bone ash, 3% of the measurement represents fat, and 8% of the measurement represents protein and water.

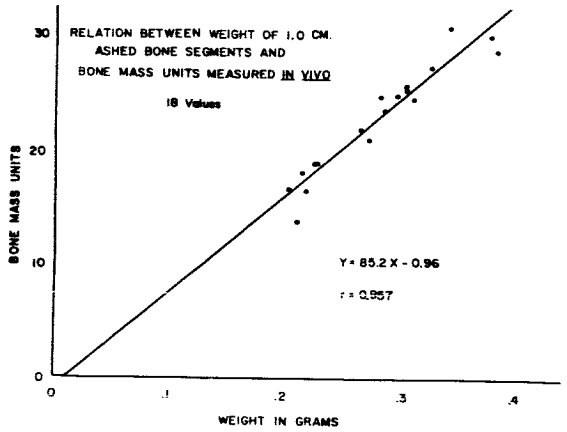

FIGURE 3.—Comparison of bone mass measurements made *in vivo* with weight of ashed 1-cm bone segments.

QUESTION OF ARTIFACT AND SENSITIVITY

The possibility that this measurement represents an artifact, or at least a parameter that does not vary in a biologic setting, seems unlikely but was evaluated by making sequential measurements in a group of eight pullets. Five pullets were placed on a low calcium (0.1% calcium but otherwise nutritionally adequate) diet, and the other three hens which served as controls were permitted oyster shell *ad libitum*. As shown in figure 4, the bone mass, shown as means for each group, declined in the group deficient in dietary calcium until the 15th day when the curve "leveled off." This "leveling off" was accompanied by cessation of egg production. The control group did not change in bone mass. This change in bone mass is in agreement with measured changes in total carcass calcium by other workers and lends support to the conviction that the measurement of bone mass is a reflection of total body calcium. By plotting the number of eggs laid against bone mass (as shown in fig. 5), a number of regression curves can be obtained. Inspection of these curves indicates that, in the main, this technique is capable of measuring the bone mineral loss following laying a single egg. In this study the average decline in bone mass with each egg laid was 0.76 bone mass unit or equivalent to 8.7 mg bone ash per linear centimeter of the bone measured. If the total body cal-

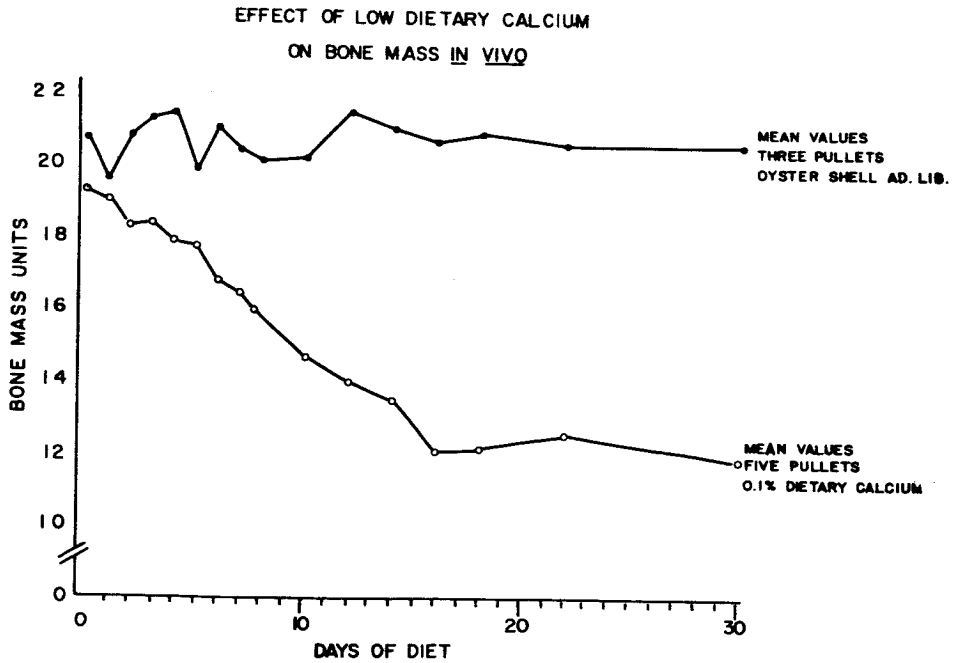

FIGURE 4.—Bone mass changes in pullets with severely restricted dietary calcium.

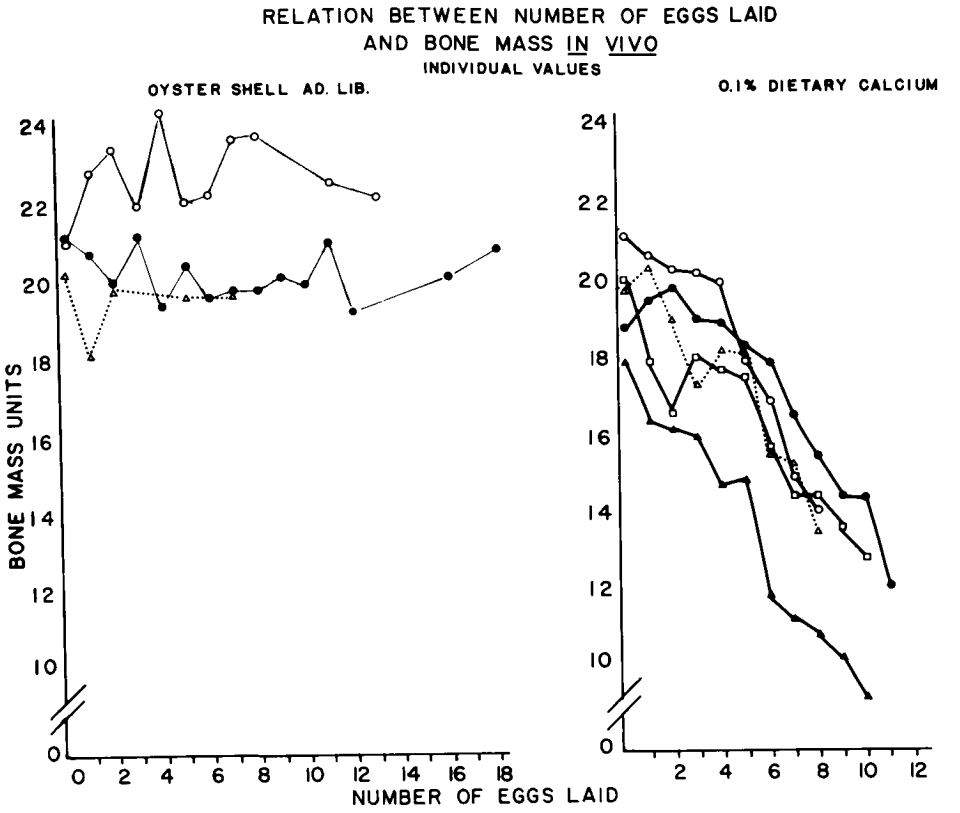

FIGURE 5.—Change in bone mass in pullets with each egg laid, when receiving a low calcium diet.

cium is about 21 grams, each egg laid represents a loss of body calcium of about 10%.

CONCLUSION AND SUMMARY

The measurement of bone mineral content by the Cameron-Sorenson technique of attenuation of a mono-energetic photon beam is an accurate measure of bone weight, or mass, that is reproducible within at least 5% and capable of detecting a change in total body calcium of the order of 10% under the conditions of these studies.

REFERENCES

CAMERON, J. R.; and SORENSON, J.: Measurement of Bone Mineral *in vivo:* An Improved Method. Science, vol. 142, 1963, pp. 230–232.

MAZESS, R. B.; CAMERON, J. R.; O'CONNOR, R.; and KNUTZEN, D.: Accuracy of Bone Mineral Measurement. Science, vol. 145, 1964, pp. 388–389.

CAMERON, J. R.: Bone Mass Determination Using Monochromatic X-rays and a Scintillation Detector. Presented at current symposium.

Part II

ACUTE CHANGE IN ACTIVITY

The relation between stress and strain acting on bones and its importance to calcium balance has been recognized. Hens raised in layer cages tend to develop thin, frail bones which decrease the market value of the hen when slaughtered. It has been suggested that the hen raised in layer cages develops demineralized bones as a consequence of the limited activity associated with relatively restricted quarters. In an attempt to demonstrate an effect of the change in activity on bone mass, ten mature hens, all of which had been raised in layer cages, were divided into two groups. The first group remained in the cages and served as controls. The second group was transferred to a large (10 x 12 feet) pen which had been fitted with a wire floor to prevent access to litter. All hens received the same diet (a normal layer diet with oyster shell) and lighting conditions throughout the experiment; and these were unchanged from prior to study. Both groups were weighed, and bone mineral measurements, according to the method previously described, were made at the start and after 14 days. The results are shown in figure 1. Individual changes are shown with group averages represented by dotted lines. Average bone mass for the caged group was 24.8 units at the start of the study and 25.0 at the end; it was 23.5 for the group placed on the floor at the start and the exact same value at the end of 14 days of increased activity. While average body weight of the caged group showed a slight increase, it decreased for the group on the floor. Therefore, no change in bone mineral content was demonstrated with an increase in activity of 14 days' duration.

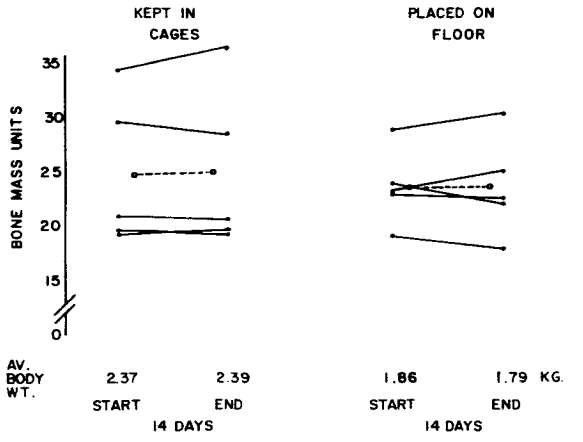

FIGURE 1.— Bone mass measurement before and after acute change in activity. Group means are shown with dotted lines and did not change.

BONE MINERAL CHANGE WITH MATURATION

A positive calcium balance has been demonstrated in the 2- or 3-week period which precedes laying the first egg. In an attempt to demonstrate this calcium storage, bone mineral measurements were made in three 109-day-old pullets at various intervals during their maturation. Initially, they were reared in a floor pen with a standard growth diet; after 35 days they were transferred to layer cages with a laying ration consisting of 1.5% calcium plus oyster shell. This transfer was made well before the first egg was laid. The growth curves, expressed as a percent of the first measured value, are shown is figures 2, 3, and 4. The average increase in bone mass for the three pullets was 63% and the average increase in weight was 43%. Daily egg production was 76% for pullets #46 and #47, and 90% for pullet #48 following the first egg laid. These measurements indicate that the positive calcium balance which precedes egg laying is reflected in a greater increase in bone mass than can be accounted for by the increase in body weight. It also appears that there are abrupt fluctuations in the bone mineral content which precede by several days the first egg laid.

NUTRITIONAL CALCIUM REQUIREMENT

The optimum calcium supplement for laying hens has been estimated at 2.75% by the National Research Council (1962). This requirement has been based principally upon parameters of egg production and shell thickness. The prob-

lem of thin, frail bones which fracture during processing raises the possibility that the rec-

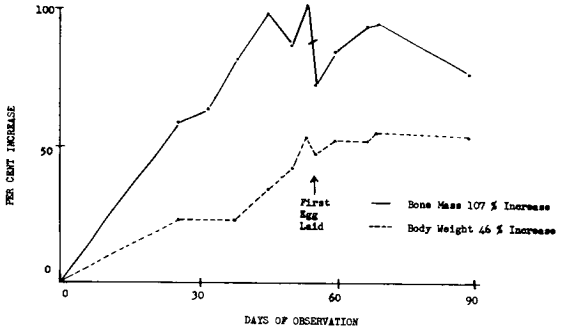

FIGURE 2.—Changes in bone mass and body weight (expressed as a percent of the initial measurement) with sexual maturation.

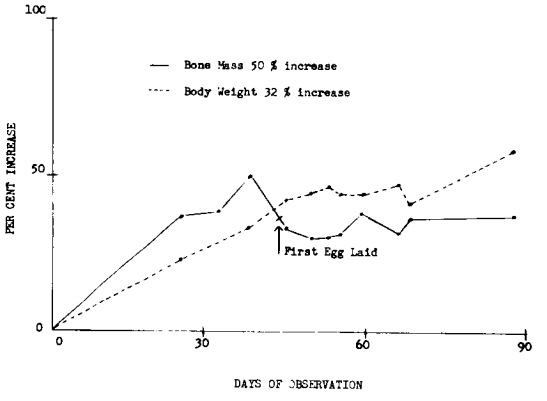

FIGURE 3.—Changes in bone mass and body weight (expressed as a percent of the initial measurement) with sexual maturation.

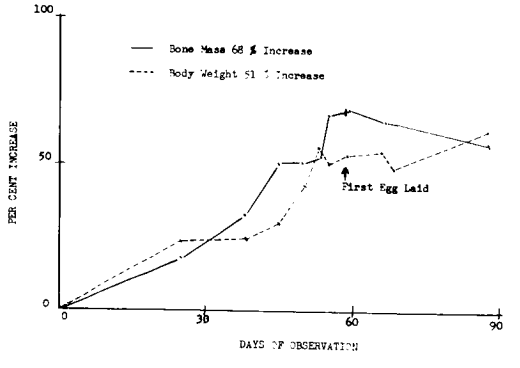

FIGURE 4.—Changes in bone mass and body weight (expressed as a percent of the initial measurement) with sexual maturation.

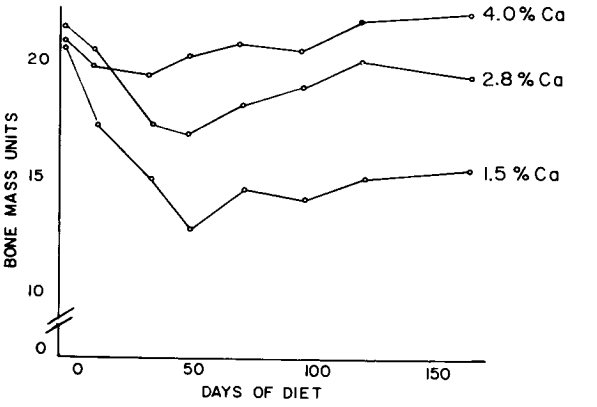

FIGURE 5.—Bone mass changes in three groups of ten hens each with different levels of dietary calcium.

ommended supplement of calcium may not be adequate to maintain bone strength. To investigate this possibility, 40 adult hens were selected and divided into four groups according to body weight and egg production records. Each group received diets containing different amounts of calcium. Bone mineral measurements, body weight, and egg production were recorded at various intervals. Except for calcium, the diets were nutritionally adequate; the phosphorous content was 0.74% for all groups. The change in bone mass with duration of the various diets is shown in figure 5. The data points represent averaged values for the ten hens in each group. The fourth group, which received a lower phosphorous ration, is not shown. The data show a decline in bone mass for the group which received 2.8% calcium in contrast with the group receiving 4.0% calcium. Egg production was comparable for these two groups, and was greatest just prior to the measurement at the 52nd day of diet. The regression curve "levels off" at this point as the egg production declines. Customarily, first-year laying hens are slaughtered shortly after this decline in egg production. Figure 6 shows the marked decline in bone mass for two individual hens selected from the group receiving 2.8% calcium. It can be seen that although a 4.0% calcium level will adequately maintain bone mass, the 2.8% level does not. Thus, if this parameter is considered, optimal calcium supplemental for the laying hen may be higher than the currently recommended 2.75%.

CAGE LAYER FATIGUE SYNDROME

This disorder, sometimes called "cage layer osteoporosis," is characterized by leg weakness,

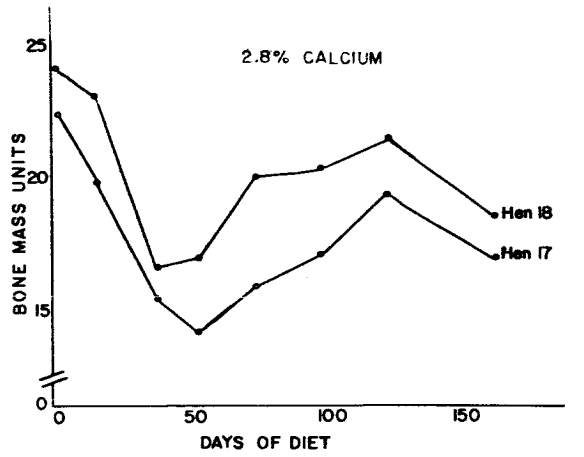

FIGURE 6.—Bone mass changes in two hens receiving 2.8% dietary calcium.

demineralized bones, normal serum calcium, phosphorous, and magnesium levels, and the absence of other recognized causes of leg weakness; it occurs in hens raised in layer cages. The etiology and pathologic physiology is controversial or obscure. Hens with this condition were sent to the University of Wisconsin Poultry Science Laboratory from interested poultry raisers in the vicinity. Figures 7 and 8 show sequential bone mass measurements in six of these animals. Those in figure 7 were placed in a floor pen with wood shavings while those in figure 8 were separated from the litter by being placed in a large cage with a wire floor. All the animals were in poor condition upon arrival and were unable to walk. The hens 1, 3, and 6 were able to walk within 3 days and were laying by 12 days of observation. Hens 2, 4, and 5 were able to walk after 17 days and were laying after 23 days. These birds received a standard laying diet with oyster shell supplementation, and food and water were placed in individual containers near the disabled animal. The initial bone mass measurements of these hens were all lower than the normal range (17 to 25 bone mass units, not corrected for age or body weight), and the general improvement correlated with increase in bone mass values. This general correlation was also noted in two birds which developed the cage fatigue syndrome while under observation at the Poultry Research Laboratory and while receiving diets containing 2% calcium. It appears that this syndrome is related in some way to mineral metabolism and that this is reflected in bone mass measurements.

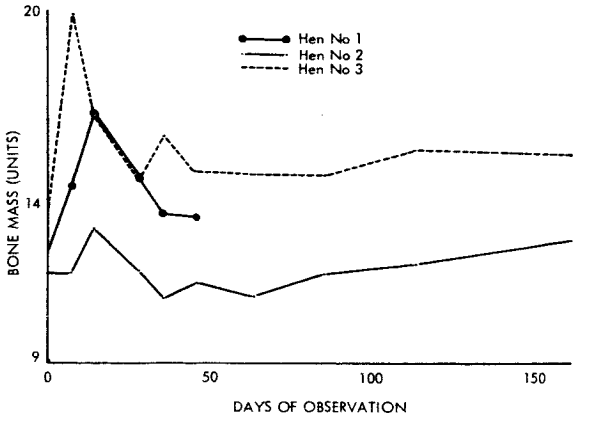

FIGURE 7.—Bone mass changes in hens recovering from cage fatigue syndrome.

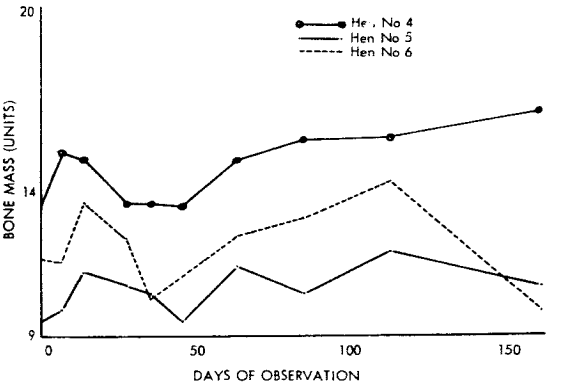

FIGURE 8.—Bone mass changes in hens recovering from cage fatigue syndrome.

These measurements of bone mineral content under various conditions should not be construed as detailed studies of the topics under consideration, but rather as examples of the application of bone mineral measurements to an animal system.

ACKNOWLEDGMENTS

The authors express their appreciation to Drs. John R. Cameron, Department of Radiology and Physics, and M. L. Sunde, Department of Poultry Science, for support and encouragement, and for making equipment and facilities

available. The authors also thank David Knutzen and Richard O'Connor for performing measurements.

COMMENTS

Dr. URIST. This brings up a third application of the problem of bone density measurements. First is the apparent problem of NASA, second is the clinical problem of osteoporosis, and third is a domestic economic problem, the problem of the laying hen.

I think Dr. Babcock's demonstration is a more severe test for bone mass measurement, I should say bone ash mineral measurement, than any I can think of in the human, because when the hen develops a loss of bone ash or ash mineral on a low calcium diet as you say, she does continue to make bone.

For the first 4 or 5 days, as a matter of fact, there is a great deal of bone matrix and insufficiently mineralized matrix. To meet the exigency of the moment, she actually makes more bone than there was before in the marrow cavity. If this method detects the ash within 4%, this testifies to the accuracy of the method because you could not possibly do it with film. You could not get a mixture of crystals in film and see differences to this extent. I have tried it with film and with chemical measurements for calcium and compared the two.

I hope that it will not be out of order to ask how much cage layer osteoporosis there is in Wisconsin.

Dr. BABCOCK. Cage layer fatigue is an important problem in all states. It is more particularly important in the warmer climates. It seems to be principally a problem of hens raised in layer cages. In this country this is an increasingly common problem.

Precision Methods Using Soft Penetrating Radiation for Bone Densitometry

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The soft X-radiation instrument possibilities in bone densitometry will be highlighted in this paper. This discussion is not intended to be a comprehensive review or a detailed forecast of the instrument possibilities in this field. We will review only those features of prior work which are significant to this discussion. Some tentative suggestions concerning instrumentation and the way these possible approaches may affect the overall research objectives are included.

Following some introductory speculations to promote informal discussion outside the scope of this paper, we will specifically discuss two related instrument approaches. One instrument uses a two-gamma radioisotope source for both ground and flying clinical studies. The other instrument uses an X-ray tube to explore the ultimate limits of laboratory precision and to evaluate the accuracy of the simpler approach. The factors relating to the precision and accuracy of measurement and related design problems of these two instrument approaches will be analyzed in as much detail as time allows.

Our discussion considers only the uses of soft radiation in a narrow beam geometry which undergoes nearly exponential attenuation. The attenuation factor is sensitive to both the atomic number of the absorbing material and the energy of the radiation in a characteristic way. These properties permit the quantitative analysis of materials. Living materials introduce the necessity for the use of lower energies for analysis.

The utilization of soft X-rays for the compositional analysis of complex materials has been proposed and carried forth by many investigators for many years (Jacobson, 1964; Jackson and Lindberg, 1964; Sorenson and Cameron, 1964). This approach and analysis has been generally characterized by the consideration of one or more monochromatic radiation beams in a well-collimated, narrow-beam geometry (Jackson and Lindberg, 1964; Lefker, 1964).

The virtue of monochromaticity and collimation is the resulting simplicity in the mathematical analysis.

Instrumentation at the 2% precision level has already been used in the laboratory environment (Cameron and Sorenson, 1963). Errors arising from problems of sample motion, characterization of the model, and others may limit the accuracy to the 3% or 5% level.

There is naturally a desire to improve the precision and accuracy of measurement in bone densitometry to better than 1% and then on to the 0.1% level. Further, there is a need for reliable flying equipment outside of the ground laboratory environment. Such instrument improvement can be used to measure small rates of change of the bone densitometry status.

In the unexplored instrument design territory, a view of the over-all situation is given in table I. There are elements of pure speculation, considerable obscured vision, and not a little courage involved in offering this brief summary for your consideration. Shown are promising areas of further study to quantify

TABLE I.—*Soft Radiation Counting—The Overall Abbreviated View*

Radiation source and detector	Energy spectral properties	Source properties	Applicable theoretical analysis	Instrument sensor properties	Percent accuracy range	Project time frame	Current status
Radioisotope	Narrow band.*	Thin sources. Low activity	Elementary.	----------	±5 to 10	------	----------
			----------	----------	--------	------	----------
and	Many narrow bands.**	Thin sources. Low activity	Simple----	Portable---	±0.5 to 1.	1 yr--	Understood.
			----------	Flyable---	--------	------	----------
NaI(Tl) detector.	Broad band.#	Thick sources. Higher activity.	Complex--	Portable---	±0.2 to ±0.5.	1–2 yr-	Unexplored.
			----------	Flyable----	--------	------	----------
Tube-target	Narrow band.*	Poor quantum efficiency.	Elementary.	----------	--------	------	----------
and	Many narrow bands.**	Large machines. Inefficient---	Simple----	Heavy----	±0.05 to 0.1.	1 yr---	Understood.
			----------	----------	--------	------	----------
NaI(Tl) detector.	Broad band.##	Efficient-----	Complex--	Transportable. Portable.	0.1-----	2–3 yr-	Unexplored.
Ion chamber detector.	Broad band.###	Programmed volt. Efficient ----	Complex--	Flyable---	0.5 to 1--	2–3 yr-	Unexplored.
			----------	----------	--------	------	----------

*Narrow band is a monochromatic energy band.
**Many narrow bands are polychromatic, i.e., several narrow bands used sequentially.
#Broad band is many narrow contiguous bands used simultaneously.
##Broad band is many narrow bands used sequentially.
###Broad band is sequential use of different ## broad bands obtained by programmed voltage application of the tube source.

some very rough estimates set forth for both the ground and flying equipment capability.

In the actual world of soft radiation sources, especially where strong, small sources are required, these sources are not truly monochromatic.

The strong radioisotope source is a thick source with degraded radiation due to self-absorption. Other energies may also be present.

X-ray sources are also naturally broad band. An effort to make the radiation monochromatic with filters requires that considerable amounts of radiation be thrown away. Although precision measurements can be made in the 0.1% range by starting with a higher power machine, one naturally asks the question, "Why fight it?"

Instrumentation planning must consider methods of using existing broad band sources to achieve efficient use of the generated radiation quanta. In the analysis of data from broad band sources, computers will be necessary to obtain the answers because the analysis is necessarily more complex. In reality, the current approach to measurement is a simplification of the actual problem to the point where the elementary analysis can be handled by a slide rule. This simplification is a significant source of loss of accuracy and inefficiency.

In practical instrument design, the factors of reliability, size, and weight become as important as accuracy from the viewpoint of practical utilization. This is particularly true where

flying equipment is concerned. Maximum bone densitometry information from each pound of equipment and from each watt of power is needed. Both minimum and safe radiation exposure is another ingredient of design.

A practical objective from this efficiency viewpoint is to use each quantum originating from either the radioisotope or tube source. The longer term instrument design direction must utilize computational techniques as yet not worked out. These future techniques will use the quanta presently filtered out to permit the use of the simpler, slide rule mathematical analysis. For this reason broad band radiations are included for its future potential in table I. From a mathematical viewpoint, the broad band radiation beam may be considered as the simultaneous presentation to the sample of collimated radiation of adjacent monochromatic energy bands. This is termed simultaneous polychromatic radiation. This is in contrast to the method of sequential use of different monochromatic radiations, the analysis of which leads to the n simultaneous equations which can be solved explicitly for n elements (Jackson and Cameron, 1964; Lefker, 1964).

In the special case of a tube X-ray source, there is the capability of varying the electron accelerating potential. This capability permits a method of obtaining a variety of sequential polychromatic radiation sources. In this method the differences between successively emitted broad band spectra, as the voltage on the tube is decreased in increments, can be utilized for an analysis procedure. The capability of this method is certainly worth further study.

As we return to more trodden paths, the sequential polychromatic tube X-ray source is a proper point of immediate departure. The use of several monochromatic radiation sources in sequence generated by heavy filtration of an X-ray tube offers the practical opportunity to explore and determine the accuracy limitations in bone densitometry.

Aside from its clinical virtues, a tube X-ray source of this type evaluates the residual accuracy assumptions of simpler instruments. Such a simpler portable and flyable instrument may be built using an alternating dual chromatic radioisotope instrument such as using I^{125} and Am^{241}. We consider both of these specific instrument approaches and their interrelationship to improve the accuracy of measurement.

PRECISION AND ACCURACY

A digression is appropriate to discuss precision and accuracy. For our purposes here, precision is a measure of the repeatability of the instrument on a given sample which apparently doesn't change, such as a phantom.

Instrumental accuracy, on the other hand, is a measure of the truth. How closely does the precision measure correspond to the actual state of affairs? In a living sample the accuracy of even a precise instrument is limited by its design characteristics. It can only measure according to the instructions it receives. It interprets the sample according to a model of the living sample and its inert interpretation, the phantom.

MODELS AND PHANTOMS

The accuracy of the instrument is meaningful for a quantitative understanding of the living sample. The living sample is replaced in the mind of the instrument designer and the instrument user with a model. In the calibration and test of the instrument, he uses a phantom design based on these models. Typically, the models are here classed as either a physical element model or as a biological material model (table II). The physical element model treats the elements as independent variables. The biological model treats the biological and metabolic materials as independent variables from a functional point of view.

THE DUAL MONOCHROMATIC AND N-MONOCHROMATIC DESIGN

The tube X-ray method utilizes the physical model of the sample, for example, the heel bone (os calcis). The physical model treats the elements as seven independent variables.

In the simpler dual gamma instrument, the assumptions are made about the relationship between the two models of the structure. These

TABLE II.—*The Sample Under Measurement, Its Equivalent Model*

	Model	Characteristics	Examples	Application area
The measured *in vivo* sample.	Physical	Elements	H, C, N, O, Ca, P, others.	Preferred for mathematic and analytic treatment.
	Biological	Metabolic and biological materials.	Hydroxyapatite, water, fat, muscle, protein, others.	Preferred for clinical understanding.

Physical—The 2-γ analysis requires assumptions to correlate dynamics of physical and biological models.
Biological—The n-γ sequential polychromatic method requires no assumptions concerning the model dynamics.

assumptions introduce errors which still require quantitative evaluation.

Herein lies the significant virtue of the larger tube machine. This machine can be operated in many instrument modes from dual to n-polychromatic to experimentally valuate the effects of model assumptions in the actual clinical situation.

QUANTITATIVE PRECISION AND ACCURACY ANALYSIS

The use of a precision laboratory X-radiation source of n sequential monochromatic radiations will permit the analysis of living bone material under conditions which permit the application of the simplest theory of the bone densitometry analysis. This theory utilizes the physical model of n elements. With adequate radiation and the precision features of geometrical stability and radiation stability, the opportunity exists for substantial clarification of mineral and nonmineral metabolic activity.

This knowledge can, in turn, be used to clarify the type of correction needed to improve the accuracy of a simpler dual gamma instrument approach. Knowing the magnitude of the errors introduced in the simpler equipment will naturally eliminate uncertainty and improve confidence in its accuracy level.

The main sources of errors are listed here. The discussion below treats them to a greater or lesser extent in conjunction with an example of the os calcis measurement and the complementary two-instrument approach. Errors that are principally those of precision are statistical fluctuations, monochromaticity assumptions, and collimation and scattering. Errors that are either systematic or instrumental and affect accuracy relate to the validity of model, including physical elements, biological materials, and the motion of sample.

THE SEVEN-ELEMENT X-RAY TUBE METHOD

The mathematical treatment of the sequential polychromatic approach consists of considering that the model of an *in vivo* bone consists of n elements, typically H, C. N, O, Ca, P, S, and other trace units. With seven monochromatic energies sequentially irradiating the sample, the amount of each element can be determined.

The typical family of equations consists of

$$ {}_1I = {}_1I_o \exp -({}_1\mu_1 X_1 + {}_1\mu_2 X_2 + \cdots + {}_1\mu_7 X_7) \quad (1) $$

$$ \vdots $$

$$ {}_7I = {}_7I_o \exp -({}_7\mu_1 X_1 + {}_7\mu_2 X_2 + \cdots + {}_7\mu_7 X_7) \quad (2) $$

Here the right subscript refers to the element in the sample. The left subscript refers to the energy. Thus, ${}_1I_o$ is the unabsorbed intensity of energy 1. X refers to the thickness of the element of the right subscript in grams per square centimeter. The mass absorption coefficient μ in square centimenters per gram applies to the energy of the left subscript and the material of the right subscript.

The measured attenuation factors ${}_1I/{}_1I_o$ of the seven energies is related to seven elements and seven known mass absorption coefficients for these elements. Typically, this 7×7 equation matrix can be solved using a computer for each element. The precision of measurement

can be worked out as a function of source strength, mass absorption coefficient values and their differences, and the other purely mathematical and radiation statistical fluctuations. The mathematics is simple to a programmed computer.

The advantage of this approach is that, from a clinical point of view, no assumptions are made beyond the fact that each element may experience transport in or out, independently.

A metabolic change in the status of the sample is independently evaluated of the actual biological chemicals which may undergo transport. Certainly the over-all changes in the element ratios may be explained by certain amounts of bone mineral, water, fat, muscle, or other substances. The many possibilities are not part of the instrument measurement or model assumptions.

For these reasons the seven-gamma method presents the opportunity for measurement with the greatest accuracy. We now reduce the number of independent variables by grouping them into two natural groups and use two radiations for measurement. The rationale of this procedure is that there is naturally a heavy element group 1—bone minerals (Ca and P)—and a light element group 2—flesh material (H, CNO). The question arises that perhaps three groups are better, i.e., to separate H and C, N, O. This is not answered as yet. This question is resolved from an experimental point of view by the fact that two radiations are readily available from radioisotopes, namely, I^{125} and Am^{241}. Further, the X-ray instrument can be operated in a 2- or 3-radiation mode to experimentally establish the accuracy of the simpler approach by comparing it with the 7-radiation answer on the same sample. Still using the physical model, we now reduce the number of monochromatic radiations to two arising either from the X-ray tube or from I^{125} and Am^{241}. Here now we can compare two instruments.

THE TWO-ELEMENT GROUP-DUAL RADIATION METHOD

In the composite sample, such as the os calcis, a model consisting of two distinct groups of elements is considered to comprise the total sample thickness. The thickness is expressed in mass absorption units of grams per square centimeter at the measurement location (Cameron and Sorenson, 1963; Grodstein, 1957). At this location two monochromatic radiation beams in a well-collimated beam undergo exponential attenuation described by

$$ {}_aI = {}_aI_o \exp\ (-{}_a\mu_1 X_1 + {}_a\mu_2 X_2) \qquad (3)$$

$$ {}_bI = {}_bI_o \exp\ (-{}_b\mu_1 X_1 + {}_b\mu_2 X_2) \qquad (4)$$

The subscripts a and b refer to the energy, and the subscripts 1 and 2 refer to the element groupings. Typically, material 1 is calcium and phosphorus content, and material 2 is the hydrogen, carbon, oxygen, and nitrogen content of the sample.

In a typical bone demineralization situation, where the sensitivity of the method is considered, the bone elements depleted are relatively small. The amount of bone elements removed is ΔX_1. Associated with this loss is a corresponding loss of the other elements H, C, N, O associated chemically with Ca and P. Additionally, other metabolic changes may independently add or subtract to the H, C, N, O content. Thus, the value of ΔX_2 is not necessarily functionally related to ΔX_1.

Precision and Accuracy of Measurement

The basic two-equation approximation to the bone model is now explicitly solved for the thickness and precision relationships. The function R_a, R_b is defined as the logarithmic ratio of the linear attenuation factor given as follows:

$$R_a = ln \frac{{}^aI_o}{{}_aI} = {}_a\mu_1 X_1 + {}_a\mu_2 X_2 \qquad (5)$$

$$R_b = ln \frac{{}^bI_o}{{}_bI} = {}_b\mu_1 X_1 + {}_b\mu_2 X_2 \qquad (6)$$

From this pair of simultaneous linear equations, the value of X_1, the thickness of the mineral elements, can be explicitly expressed as

$$X_1 = \frac{R_{ab}\mu_2 - R_{ba}\mu_2}{{}_a\mu_1{}_b\mu_2 - {}_a\mu_2{}_b\mu_1} \qquad (7)$$

A similar expression for the thickness of the non-mineral elements is given by

$$X_2=\frac{R_{ab}\mu_1-R_{ba}\mu_1}{{}_a\mu_{1b}\mu_2-{}_a\mu_{2b}\mu_1} \tag{8}$$

From these expressions, the effect on precision of measurement arising from the statistic fluctuations of counting can be derived. The statistical uncertainty arising from the two independent counting fluctuation errors is given by the standard general expression. This expression, using the nomenclature of this discussion, may be used to calculate the fractional deviation to be expected in X_1, the standard deviation measure of the fluctuation of the total count used in the measurement.

$$\left(\frac{\Delta X_1}{X_1}\right)^2=\left(\frac{1}{X_1}\right)^2\left\{\left(\frac{\delta X_1}{\delta_a I}\right)^2(\sqrt{{}_aI})^2+\left(\frac{\delta X_1}{\delta_b I}\right)^2(\sqrt{{}_bI})^2\right\} \tag{9}$$

Here, $\frac{\Delta X_1}{X_1}$ is the fractional deviation in X_1 expected to arise from the standard deviation fluctuation, $\sqrt{{}_aI}$ and $\sqrt{{}_bI}$, expected in the measured count ${}_aI$ and ${}_bI$.

Utilizing equation (7), the partial differentiations are performed and followed by the algebraic manipulations indicated. The following result for the fractional error is obtained

$$\frac{\Delta X_1}{X_1}=\left(\frac{1}{R_{ab}\mu_2-R_{ba}\mu_2}\right)\left(\frac{{}_b\mu_2^2}{{}_aI}+\frac{{}_a\mu_2^2}{{}_bI}\right)^{1/2} \tag{10}$$

This expression can be rewritten in terms of the unabsorbed source count

$$\frac{\Delta X_1}{X_1}=\left(\frac{1}{R_{ab}\mu_2-R_{ba}\mu_2}\right)\left(\frac{{}_b\mu_2^2e^{R_a}}{{}_aI_0}+\frac{{}_a\mu_2^2e^{R_b}}{{}_bI_0}\right)^{1/2} \tag{11}$$

For equal contributions from source a and source b to the error, the equality of the following terms holds

$$\frac{{}_b\mu_2^2e^{R_a}}{{}_aI_0}=\frac{{}_a\mu_2^2e^{R_b}}{{}_bI_0} \tag{12}$$

Rearranged, then, the source strengths for equal time of measurements bear the following ratio

$$\frac{{}_aI_0}{{}_bI_0}=\frac{{}_b\mu_2^2e^{(R_a-R_b)}}{{}_a\mu_2^2} \tag{13}$$

Thus, for the precision of measurement relationship, equation (11) can be expressed in terms of the lower energy a on the basis of equal contribution to the error using equation (13) as follows:

$$\frac{\Delta X_1}{X_1}=\frac{b\mu 2^{R_a/2}}{R_{ab}\mu_2-R_{ba}\mu_2}\sqrt{\frac{2}{a^Io}} \tag{14}$$

We can now illustrate the use and limitations of the equation with the example of the os calcis measurement.

Os Calcis Physical Element Model

For the purpose of numerical evaluation of the two-gamma method, a model of the os calcis is set up. This model consists of 2 cm of tissue, 2 cm of tissue matrix within the bone, and 2 cm of dense bone of 1.85 g/cm³ (fig. 1). The dense bone portion is considered, in turn, to be composed of approximately 30% weight of the calcium and phosphorus elements. Thus, the total sample is composed of two fractions according to the physical element model. The Ca, P (fraction 1) is (2 cm×1.85 g/cm³)×30%, which is 1.1 g/cm². This is a typical value for X_1. The H, C, O, N (fraction 2) is 4 cm×1.0 g/cm³ and (2×1.85 g/cm³)×70%, which is 6.6 g/cm². This is a typical value for X_2.

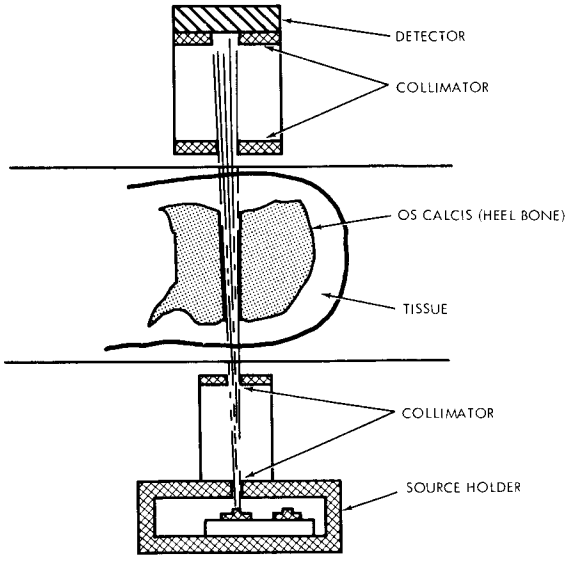

FIGURE 1.—Source and beam geometry dual gamma os calcis management.

Experimental Arrangement

The radioisotope sources are 100 mCi of I^{125} and 1 mCi of Am^{241}, alternately placed in front of the aperture. The source geometry consists of a source diameter of 2 mCi. The beam is defined by collimation to produce a beam of 2 mm diameter at 10 cm from the source. The exit collimator of the source is a dual aperture to avoid unnecessary exposure and production of scattered radiation. The entrance collimator of the NaI (T1) detector preserves this aperture with a dual aperture arrangement to avoid the detection of scattered radiation.

Radiation Sources and Monochromaticity

The radiation from I^{125} is reported to be principally 27.5 keV, with the remainder 31 keV (15%), 35 keV (5% to 7%) (Sorenson and Cameron, 1964). This mixed radiation can be filtered to increase the relative proportion of the 27.5 keV radiation or used unfiltered. In either case, a correction can be applied to the measurement based either on a calibration procedure or a mathematical treatment. Such a correction is not considered here but is necessary in thick samples, where the absorption effects of the 27.5 and 35 keV radiation become appreciable.

Absorption Coefficients

Typically, for the purposes of the present example, the representative values of the mass

TABLE III.—*Representative Values of Mass Absorption Coefficient Os Calcis (physical elements model)*

Physical elements model	I^{125} energy *a*, cm²/g	Am^{241} energy *b*, cm²/g
(1) Ca, P	$4.8={}_a\mu_1$	$0.19={}_b\mu_1$
(2) H, C, N, O	$0.3={}_a\mu_2$	$0.54={}_b\mu_2$

absorption coefficients have been estimated and listed in table III. The mass absorption coefficient for the calcium-phosphorus component is based on an equal weight ratio of calcium to phosphorus in the material lost from the bone.

Statistical Fluctuation Error-Numerical Calculation

The estimate of the contribution of the error from statistical fluctuations is made from the analysis above.

First, the value of the R is calculated. From equation (5)

$$R_a=4.8\,\frac{\text{cm}^2}{\text{g}}\times 1.1\,\frac{\text{g}}{\text{cm}^2}+.30\,\frac{\text{cm}^2}{\text{g}}\times 6.6\,\frac{\text{g}}{\text{cm}^2}=7.3 \tag{15}$$

From equation (6)

$$R_b=.19\,\frac{\text{cm}^2}{\text{g}}\times 1.1\,\frac{\text{g}}{\text{cm}^2}+.54\,\frac{\text{cm}^2}{\text{g}}\times 6.6\,\frac{\text{g}}{\text{cm}^2}=3.8 \tag{16}$$

Using these numerical values and the mass absorption coefficient values of Table III, the following relationship, equation (17), for the fractional error in mineral elements as a function of the unabsorbed count measured at the detector is found from equation (14). The contribution to the error from each source is considered equal.

$$\frac{\Delta X_1}{X_1}=\frac{10.6}{\sqrt{{}_aI_0}} \tag{17}$$

Thus, for an error of $\pm 1\%$ under the conditions of this problem, the 27.5 keV source *a*, produces

$${}_aI_0=\left(\frac{X_1}{\Delta X_1}\right)^2(10.6)^2=10^4\times(10.6)^2$$

$\cong 1.2\times 10^6$ counts without the sample present.

An equal part of this error arises from the 59.6 keV, source *b*, whose strength from equation (13) is

$${}_bI_o=\frac{a\mu_2^2}{b\mu_2^2}\,e^{\frac{{}_aI_o}{(R_a-R_b)}}=\left(\frac{0.3}{0.54}\right)^2\frac{{}_aI_o}{e^{7.3-3.8}}$$
$$=9.3\times 10^{-3}\,{}_aI_o\cong 1.2\times 10^4 \text{ counts}$$

Thus, the count in the detector from the higher energy source *b* need only be about 1/100 the

count from the lower energy source a for equal contribution to the total error.

Error With 100 Millicuries I^{125} and 1 Millicurie Am^{241}

On the basis of the experimental geometry, the os calcis model and source strengths of 100 mci I^{125} and 1 mci Am^{241}, the error in bone mineral element depletion can be estimated from the analysis. Using equation (15), the standard deviation arising from fluctuation in measured count can be evaluated from the unabsorbed count rate expected.

The unabsorbed count from I^{125} is given by

$$ {}_aI_o = 3.7\times10^7 \text{ mc } \frac{d}{\text{sec}} \times 100 \text{ mc} \times 1.4 \frac{\gamma}{d} \times \frac{\pi(0.1)^2 \text{ cm}^2}{4\pi(10)^2 \text{ cm}^2} = 1.3\times10^5 \frac{\gamma}{\text{sec}} \quad (18) $$

Thus, using equation (15)

$$ \frac{\Delta X_1}{X_1} = \frac{10.6}{\sqrt{{}_aI_o}} = 0.03 \text{ per second count} \quad (19) $$

For a 9-second count each from both sources, the error is $\pm1\%$. If a 10-mci Am^{241} source is used, the fluctuation error is reduced to $\pm0.8\%$. In table IV a summary of the fluctuation error as a function of source strength is given. For a 400-mci I^{125} source and 40-mci Am^{241} source, the fluctuation error may be expected to be less than $\pm0.5\%$.

OS CALCIS—DOSE ESTIMATE

Considerations of the radiation dose are important. The maximum skin dose may be related to the measurment accuracy under the conditions of measurement. The basis of skin dose is $9\times10^9\gamma/\text{cm}^2$ per rad at 27.5 keV and $1.1\times10^{10}\gamma/\text{cm}^2$ per rad at 60 keV (5). For a $\pm1\%$ experiment, the total count is $1.3\times10^5\gamma/\text{sec} \times 10$ sec over an area of $\pi\ (0.1)^2\ \text{cm}^2$. This is equivalent to $4.1\times10^7\ \gamma/\text{cm}^2$ from I^{125} which is approximately 0.05 rad per measurement. The dose contribution from the Am^{241} exposure is less, since only 1.2×10^4 unabsorbed gammas are incident per measurement. The Am^{241} dose contribution is about 3.5×10^{-5} rad, which is negligible. Thus, the tissue dose is due almost entirely to the I^{125}. As the error decreases and as the square root of the count and the dose increases linearly with the count, a measurement of $\pm0.4\%$ accuracy produces an I^{125} exposure of approximately 0.27 rads to the skin in the proposed experimental configuration. The tube source of X-ray will be able to supply radiation at an equivalent of $10^8\gamma$ and produces an exposure of 5 rad at 27.5 keV.

TABLE IV.—*Source Emission, Precision, and Exposure (sample spot diameter 0.2)*

Source radioisotope				Fluctuation precision level (percent)	Radiation exposure (rad)
Iodine 125		Americium 241			
mc	counts	mc	counts		
100	1.2×10^6	1	10^4	±1	0.05
400	4.8×10^6	4	4×10^4	±0.5	.051
100	1.2×10^6	10	10^5	±0.8	.24
400	4×10^6	40	4×10^5	±0.4	.27
Tube					
	10^8		10^6	±0.1	5
	10^8		10^8	±0.05	10

FLESH MEASUREMENT

In the two-gamma measurement, the opportunity exists to scan the heel. A measurement of the tissue area to the rear of the os calcis can be made. The value of this measurement of the tissue changes to bone densitometry remains to be assayed. Such a measurement after correlation with the more precise n gamma tube measurement offers a possible opportunity to correct for a possible large change of tissue status in prolonged measurements of bone demineralization.

REDUCTION OF ERRORS DUE TO MOTION OF SAMPLE

A prominent source of inaccuracy in a live sample is the serious problem of geometrical stability. This is especially true where a measurement extends over an appreciable period of time. Not only must a measurement be made at a specific spot, but also the spot must be located at some future time for a new measurement. To overcome the difficulty of landmark location, an area-scan procedure can be used. For precision measurement, motion during the scanning period results in blurring of the internal reference marks. One solution is to use a region where a well-defined beam can be moved around without changing the answer, that is, the bone is reasonably uniform in thickness.

There are instrumentation methods to correct for this motion error to any degree desired. The basic approach is to use a geometry sensor which measures the displacement of the bone densitometry sensor from its standard position. This geometrical displacement can be sensed and used to reject the densitometry data. Thus, the counting data is only registered for the time the sample is in position, i.e., the measurement is made in live-time.

The elegant approach for a geometry sensor is to use another radiation sensor to relate the measurement to an internal position where a steep gradient of density exists, such as the edge of the bone. The difficulty with this type of internal landmark geometry sensor is that these radiation beams have the normal statistical fluctuation problem. An alternate practical approach is the use of an external landmark sensor. This sensor can detect the edge of the sample by mechanical, optical, or magnetic means. Typically, a magnetic ferric oxide stripe can be painted on the skin of the heel and detected by a magnetic sensing arrangement. Such a device can detect motion of 0.01 cm in less than 0.01 sec. In fact, it may be too sensitive. The appropriate method and sensitivity desired can be worked out for the level of accuracy needed. The opportunity to eliminate motion as a major source of error does exist.

PHANTOMS

Some mention is necessary on the phantom problem. The direction for the improvement of phantoms is the use of actual elements since precision and accuracy are influenced by small differences in absorption coefficients. The elements are necessary for calibration of the mass absorption coefficients on the beam used. Compounds of these elements are needed to simulate the biological model and its dynamics.

Measurement of change of status can be simulated in the phantom with the use of the same elemental composition. The effect of cross-interference of the simultaneous transport of the several real biological materials can be evaluated in the phantoms to evaluate both the mathematical analysis and accuracy of the simpler instruments utilizing two or three radiation bands.

TUBE X-RAY SOURCE

The tube source of X-rays offers the capability with filtered monochromatic radiation to reach the $\pm 0.1\%$ precision level. Typically, at 27.5 keV, a count of 10^8 reaches the $\pm 0.1\%$ precision level at a skin exposure level of 5 rad. For rapid measurement, the integral counting is done with an ion chamber and electrometer to avoid dead time problems. (The NaI (Tl) counter is suitable for the radioisotope source at the $\pm 1\%$ level.)

The tube X-ray source can be operated in the equivalent dual monochromatic mode to compare the performance of the tube source instrument and radioisotope instrument. Many minor sources of accuracy uncertainties relat-

ing to the phantom sample and metabolic dynamics can be evaluated. The tube X-ray instrumentation becomes, relatively speaking, the instrumental standard of reference.

CALIBRATION

Calibration procedures are obviously important for the purpose of accurate measurement. The accuracy of measurement can be made to approach the precision with a different value which relates to the accuracy of the phantom and the understanding of the biological dynamics under measurement.

From a physicist's point of view, two instruments of appreciable difference in precision level can be used in an equivalent experimental arrangement. The more precise instrument then contributes to the true evaluation of the accuracy of the less precise unit.

Each instrument approach is complementary to the other. In particular, the attempt to achieve a new level of accuracy plateau in a measurement to the $\pm 0.1\%$ level by the large tube machine will assure that the portable machine will achieve its accuracy objective levels of $\pm 1\%$ or better. The large machine will permit the quantitative evaluation of the magnitude of residual assumptions present in the portable machine. Indeed, it may eliminate certain features, which may be found to be unnecessary, to further simplify and improve the usefulness of the portable machine.

CONCLUSIONS

The capability exists to build precision instrumentation for both the ground and flying clinical use. There is a need for a multipath program approach for instrumentation which ranges from the ultimate in precision instrumentation where weight and power is not considered a limitation to the reliable portable, flyable, light-weight equipment.

The precision clinical equipment will contain a minimum of physical and clinical assumptions to permit evaluation of the approximations inherent in the design of simpler equipment.

It is not unreasonable to expect a project program of instrumentation encompassing the several groups of active investigators to achieve the objectives of accuracy of $\pm 0.1\%$ or better in tube equipment and $\pm 0.5\%$ for portable and flying equipment.

REFERENCES

CAMERON, R.; and SORENSON, JAMES A.: Measurement of Bone Mineral In Vivo: An Improved Method. Science, vol. 142, no. 3589, 1963, p. 230.

GRODSTEIN, G. W.: X-Ray Absorption and Scattering Coefficients. National Bureau of Standards 583 Suppl., 1957.

JACOBSON, BERTIL: X-Ray Spectrophotometry in vivo. Am. J. Roentgenol., vol. 91, 1964, p. 202.

JACOBSON, BERTIL; and LINDBERG, BJORN: X-Ray Spectrophotometer for Simultaneous Analysis of Several Elements. Rev. Sci. Instr., vol. 35, 1964, p. 1316.

LEFKER, ROBERT: The Composition Analysis of N Component Systems by the X-Ray Absorption Method, Technical Report No. 2457, U.S. Army Engineering Research and Development Laboratories, April 1964.

SORENSON, JAMES A.; and CAMERON, J. R.: Body Composition Determination by Differential Absorption of Monochromatic X-Rays. Symposium of Low-Energy X- and Gamma Source, Chicago, Illinois, October 1964.

COMMENTS

Dr. RICH: These very high precisions with relatively low counting rates are somewhat surprising, to me at least, and lead me to wonder whether you are expecting a very low background. My question then is: Is it practical with the sort of geometry you describe and with the amount of shielding and so forth that you are going to put into flight, to count on low background rates with these high sources, a few centimeters away, and even, indeed, low noise level, etc.? In other words, if you have a high background, you cannot get this precision with these low counts.

Dr. COHEN: You must consider that a measurement is being made here with 50 millirad and here with 5 rad. The astronaut in the background is not going to have that large an exposure.

Dr. RICH: I was not thinking so much about him as about the crystal; the receiving crystal has to be shielded very carefully here.

Dr. COHEN. It is soft radiation and you have an opportunity to use heavy Z, large Z to do it.

Dr. MALETSKOS. Radiation coming in from background is a higher content. It will produce a lot of high counting rates down in the very low regions in which you are working.

Dr. COHEN. I think you will find that this is very much higher than the expected background in terms of the local doses.

Dr. MALETSKOS. Don't worry about the radiation dose to the person. We are talking about the backgrounds for the detector.

Dr. COHEN. The dose in the detector is the measure of the count precision.

Dr. MALETSKOS. Fifty millirads represents quite a number of individual events.

Dr. COHEN. That is right. It is much larger than actual backgrounds. I think you will find that the ratio of counts in the measurement to backgrounds will be 100:1 or greater.

Let me go back to one other point. You said the total number of counts here looks small. The total number of counts is related to the actual measurement with the number of mean free paths you have through your sample. If your sample is effectively thicker, your accuracy in measurement of the sample is better than the fluctuation accuracy, in other words,

$$\frac{\Delta x_1}{x_1} = 0.3\Delta\sqrt{\frac{I_1}{I}}$$

This is why this factor can be an apparent 1% error in count rate fluctuation and yet come out with a better measurement in the precision of the thickness.

Dr. RICH. Could I ask a question about the exposure? In the slide, you indicated exposures in the neighborhood of 50 millirad. Were these per measurement?

Dr. COHEN. Yes, they were per measurement, just localized at the spot.

Dr. CAMERON. It wasn't clear what experimental measurements have been made to date. Have you used a tube with monochromatic X-rays? The second part of this question is: What is the degree of monochromaticity of the output beam?

The first question is: Experimentally, what experimental measurements have been done?

Dr. COHEN. Dr. Gilson has been working in this area for several years. I have some curves we received today from Frank Day concerning some of the measurements. We actually have not evaluated a specific phantom with regard to the precision levels here. This is my version of what I think can be done. Mr. Day working with Dr. Gilson has come up with curves that are exponentials from 60 to 20 keV just to show the nearly exponential attentuation factors. We have not built anything nor proved anything at this point.

Dr. CAMERON. I would like to make a comment in regard to the analysis of errors. We have gone through this, we have included a few other things which are not included in these relatively complex slides and they are confusing to discuss intelligibly in a short period of time and with people who are not mathematically inclined. The problem is that when you go beyond a two or three component system your errors essentially go up exponentially. When you go through the analysis of these things, it is terrifically impressive how a 0.1% error in the absorption coefficient changes your results in a three-monochromatic gamma system.

Another comment I would like to make is that the impression was given here that you needed two photons to determine the mineral content of the os calsis. If this is true, you do not know the thickness of your total bone plus tissue, assuming a two-component system with no absorption coefficient, then a single monochromatic radiation absorption experiment tells you the ratio of mineral to nonmineral.

Dr. COHEN. I am trying to get another order of magnitude of precision. I can't do it with a slide rule. What you say is correct. That is why I want to emphasize that we are going on now to the next two significant figures and this is a rough problem. You are purchasing two orders of magnitude of effort.

Dr. CAMERON. I think we are down very close to 1%. The next order of magnitude down to 0.1% will require an effort of about a hundred times.

Dr. MALETSKOS. Dr. Cohen has already indicated that is so. He has a big computer to solve the equations. This is the price you pay.

My reaction from actual experience, being partly a physicist and a biologist, is that biology sometimes can get so intractible that 1% may be just the finite limit of a biological system no matter what the physics might say, and we have to keep this in mind.

Dr. MALETSKOS. In your analysis you are controlled by the absorption coefficients which you assign to what you think is represented correctly in the system. Do you have any idea how close that is or will we tend to end up with an elegant calculation but one which may be literally uninterpretable?

Dr. COHEN. I think in the final analysis we have to get three or four significant figures and possibly even more. The only way you can do this is to put the element in your machine and measure it under the same conditions you are using to measure the sample. You have to get a pure sample of the element and make your own determination of the absorption coefficient.

Dr. CAMERON. The National Bureau of Standards which has specialized in precision measurements has done considerable work in this, and their precision is about 1% on their absorption coefficient. Moving those absorption coefficients, making them more accurate by a factor of 10, is not a trivial job. You have scattering problems, geometry, and absolute measurements. This is a formidable task.

Iodine-125 Bone Densitometry

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GENERAL RADIOLOGIC PROBLEM

From the radiologic viewpoint, severe degrees of osteoporosis or osteomalacia are readily recognized. When collapse of the vertebrae with ballooning of the intervertebral disks and marked thinning of the cortex of the long bones has occurred, with normal values of serum calcium, phosphorus, and alkaline phosphatase, the diagnosis of osteoporosis is obvious. It is sometimes very difficult to distinguish between osteomalacia and osteoporosis. In fact, many radiologists call all demineralization of bone "osteoporosis" and make no effort to differentiate. It is impossible to determine minor changes in bone mineralization either from osteoporosis or osteomalacia by viewing radiographs.

OSTEOMALACIA

Osteomalacia (by definition, "soft bones") is not as common as osteoporosis. The radiologic appearance of osteomalacia in the infant and child prior to the closure of the epiphyseal plates usually poses no problem, with the osteoid seams the definitive finding. In the adult, the appearance of osteomalacia is not as specific. The appearance of the decalcification is quite uniform and the bones usually show a lack of trabecular structure. The cortex also becomes thin. In osteomalacia, noncalcified matrix is present, so that by chemical analysis one finds a decrease in mineral content per unit volume. One of the characteristic findings in far advanced adult osteomalacia is the presence of pseudofractures. These are ribbon-like zones of decalcification extending into the bone at approximately right angles to the margin. They were first described by Looser and later by Milkman and are called infractions of bone.

OSTEOPOROSIS

Osteoporosis by definition is an abnormal porousness or rarefaction of bone by enlargement of its canals or the formation of abnormal spaces. It has generally been considered as a decrease of bony tissue in which the primary disturbance is a lack of bone matrix formation with a corresponding reduction in mineral content parallel with the loss of bone matrix. This reduction in bone mass is without any proven change in its chemical composition. The values of serum calcium, phosphorus, and alkaline phosphatase are normal or, as some investigators have found, a slightly elevated serum phosphatase level. The significance of this can be argued. Bone is not static and resorption and rebuilding occur at all times in the normal person in response to stress. Whatever the cause of osteoporosis, a decrease in matrix is the end result. The loss of bone mass in some patients is accelerated over the gradual loss of bone mass which occurs with normal aging. The radiologic appearance of the bones in osteoporosis is not very distinctive. As a rule, the trabecular structure can be seen and the bones appear abnormally radiolucent. The most important finding is the thinning of the cortex. Sub-

periosteal resorption does not occur. Pseudofractures are very uncommon (Pugh, 1954).

ETIOLOGY

The classic explanation is that osteoporosis is the result of a defect in the endocrine system, resulting from any different causes and combinations of causes (Albright et al., 1941; Urist, 1958; Urist, 1959). The endocrine theory of osteoporosis received its greatest impetus in the metabolic balance studies of Albright and Reifenstein (1948).

Nordin (1961; 1962) recently suggested that the primary cause of osteoporosis is a long-standing negative calcium balance. The loss of the bone mineral being followed by the removal of matrix thus produces a reduction of bone mass without reducing the mineral content of the residual bone. Other workers (Harrison et al., 1965), on the basis of calcium metabolism studies in patients with osteoporosis, have found an increase in calcium uptake on a high calcium diet and also noticed relief of symptoms.

For many years there has been a great need for a method of assessing quantitatively the degree of mineralization of the skeleton and a means of comparing a given patient with others and the patient with himself in the fourth dimension, which is time. The method used in everyday radiological practice is the visual assessment of the mineralization grade of the skeleton on conventional radiographs. This method is handicapped by considerable individual variations and the subjective bias of the individual examiner. One cannot reproduce his results on repeated examinations of the same material. The thickness of the cortical layer of the long bones is relatively easy to measure and it can be used as an estimate of the mineralization stage in clinical radiology. It is perhaps much more accurate than the subjective estimation of osteoporosis on the basis of lumbar spine films. However, it is not a sensitive indicator of demineralization, as it is not apparent to the eye until the loss of bone is more than 10 to 15%. The changes cannot usually be easily discerned until after a loss of 20%. The method, however, has some clinical application. Barnett and Nordin (1961) use the following method. The thickness of the medial and lateral cortices of the femoral shaft at the thickest part of the cortex is measured. The sum of these two thicknesses is divided by the total shaft diameter at the same level. The fraction multiplied by 100 gives a ratio which has been variously termed the femoral or humeral score, or the cortical index. The arithmetical mean of the cortical index is 0.55 for the humerus and 0.63 for the femur in normals. This index gives an objective and easy means of investigating the grade of osteoporosis and would be useful in large scale studies of osteoporosis. If it requires a change of 20% to be measured, it is obvious that the cortical index is not exact enough to measure fine degrees of difference or for following a given patient in the fourth dimension of time.

METHODS

The method of using transmission measurements for analyses of one sort or another has been used very often in physics. Omnell (1952) presents a good summary of the work done before 1957 on bone mineral changes *in vivo*, using a standard X-ray tube as the radiation source and photographic film as the detector.

Subsequent examples of these techniques have been the work of Nordin et al. (1962) who performed desintometry on body section radiographs of the lumbar spine and compared the density of the vertebrae to that of the intervertebral disk. Mayo (1961) made quantitative measurements of bone mineral content of the os calcis and the ulna, using a step wedge with the parts emersed in water and using film to record the changes in density. Doyle (1961) made similar measurement on the ulna, using a step wedge and a special automatic recording densitometer. To eliminate the use of film as a detector, Mayer (1960) used a scintillation counter for measuring transmission of X-rays. Vose (1968) made measurements of the transmission of X-rays through the femur, using a direct counting system. Gershon-Cohen (1958) used a gamma source with direct counting of the transmitted beam by a scintillation counter.

An instrument (Lanzl and Strandjord, 1964) using radioactive I^{125} has been devised for nondestructive testing to determine the condition of the bone mineral in the skeleton. This is accomplished by studying the transmission through a single finger bone of the radiation emanating from I^{125}. For small animal work, a rear leg is used. The smaller the bone mineral content, the higher will be the transmission of the radiation through the bone. A lower value of bone mineral content can be due to a thinner bone, a bone of lower density, or a combination of both. A change in the effective atomic number of the bone would also result in a change in the radiation transmitted through it.

Our work has been stimulated by that of Cameron and Sorenson (1963) and we have followed several of their ideas. Cameron and Sorenson state that some of the ways in which their method differs from earlier methods are: (1) The transmission of the photon beam is measured directly by counting techniques, employing a scintillation detector system; (2) the photon beam used is essentially monochromatic; (3) the photon beam and detector are well collimated; and (4) the effects of the tissue around the bone are taken into account.

In the present work, the measuring unit was designed primarily to accommodate a phalanx as a skeletal sample. In particular, the second phalanx of the third digit on the left hand was chosen, since the overlying soft tissue is at a minimum, and immobilization during measurement is easily attained. Thus, correction factors and errors due to the presence of soft tissue are at a minimum. It is true that the lumbar spine, pelvis, and os calcis show a greater degree of demineralization than the small bones of the hand. However, technical considerations make estimates of mineral content of the spine or pelvis difficult. Cameron and Sorenson have used the radius of the left arm. Here, the bone is more massive; but the overlying tissue is somewhat greater than in a finger.

Our choice of the phalanx was further influenced by the work of Virtama et al. (1960), Virtama and Mähönen (1960), and Virtama (1960) who have done considerable work in correlating the mineral content of the bones of the hand with that of the rest of the skeleton. Jackson, in 1951, made preliminary studies on phalanges, using a wedge of ox bone for comparison. Garn and his co-workers (1963) at the Fels Research Institute have investigated the mineral content of the bones of the hand. Barnett and Nordin (1961) also used the metacarpals to calculate a cortical index. In consideration of the above, we chose the finger as a means to measure mineral content.

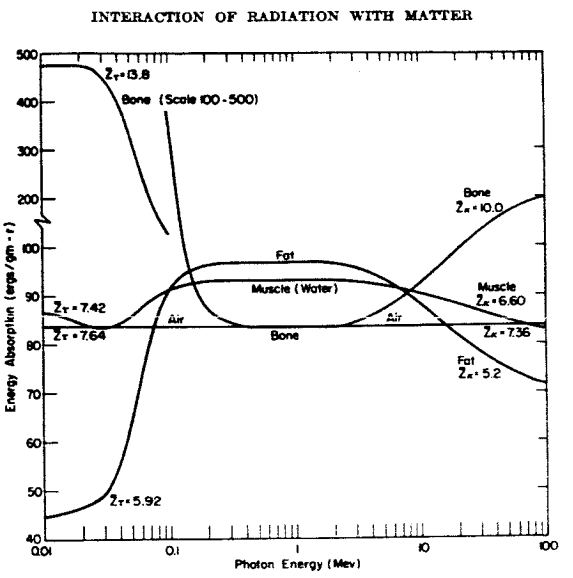

FIGURE 1.—Energy absorbed per gram-roentgen for bone, muscle, fat, and air as a function of the photon energy. From *Radiation Dosimetry* (Hine and Brownell, eds.), p. 90.

A study of the absorptive characteristics of muscle and bone tissues in humans or animals by X- or gamma-radiations reveals that the greatest discrimination occurs between 20- and 30-kilovolt energy photons (Hine and Brownell, 1956). This is shown in figure 1. Assuming an effective atomic number of 13.8 for bone, and 7.42 for muscle, the energy absorption is about 460 and 83.9 ergs/g-r for bone and muscle, respectively. From these figures, we see that the ratio of energy absorbed per gram of tissue is $460/83.9 = 5.5$ for bone to muscle. The ratio of bone to fat is still greater. When scattering characteristics are taken into account, as well as absorptive, still greater differences for bone to muscle are observed and

expected in a "good" geometry situation, which is the case for these transmission measurements.

A low energy X-ray machine can certainly produce photons in the range of 20- to 30-kilovolt energy; but, in addition, it produces many photons outside this range.

Furthermore, the intensity of an X-ray machine is not inherently stable. On the other hand, the time rate of output of a radioactive isotope of sufficiently high strength is very stable, although it does decay.

A particularly useful isotope for this purpose is I^{125}, which decays by means of electron capture to a 0.354 MeV excited state of Te^{125}. From this state, tellurium decays by means of a gamma-ray which is 80% internally converted in the K shell, giving, in turn, two X-ray photons, K_α=27 keV and K_β=31keV (Bowe and Axel, 1952). There are several other radiations such as the characteristic X-rays from the L, M, and N shells resulting from electron capture, conversion electrons, and Auger electrons; but their energy or range is such that they do not play a role in the present work.

Of the three photons, the K_α=27 keV is the most abundant, accounting for 75% of these photons per disintegration. The K_β accounts for 20% of the total, only 5% remaining as unconverted 35.4 keV gamma-ray transitions.

Consider the cross section of a finger which has been compressed between two parallel planes, V and U, as shown in figure 2. A collimated beam of X- and gamma-rays as described above is directed along path S. The transmitted intensity, I, of the beam will equal the unattenuated intensity, I_o, times the attenuation, $e^{-\mu T t}$, of the beam due to the soft tissues, where μ_T is the linear absorption coefficient in cm^{-1}, and t is the thickness of the soft tissue in centimeters. This thickness is also the distance between the parallel planes V and U. I_o is measured by simply removing the finger from its position between the two planes. From measured values of I, I_o, t and the use of expression

$$I=I_o e^{-\mu T t} \qquad (1)$$

the linear absorption coefficient, μ_T, of the soft tissue is obtained. In actual practice, it is a

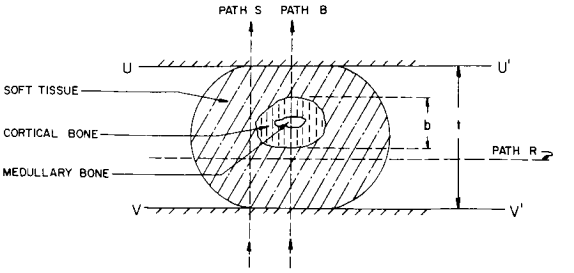

FIGURE 2.—Cross section of finger between parallel compression planes V and U.

little more reliable to rotate the finger through 90° between the planes V and U, and to make the measurements along path R.

When the beam is directed along path B, the expression contains another factor due to the absorption of the radiation in the bone. This expression is

$$I=I_o e^{-\mu T(t-b)} e^{-\mu B b} \qquad (2)$$

where μ_B is the linear absorption coefficient of bone in cm^{-1}, and b is the thickness of the bone in centimeters.

The thickness b is obtained from measurements of a radiograph of the bone in question. A magnifying lens system with a built-in reticle is used to measure the thickness. The radiograph also serves to identify the portion of the bone where the measurement is made. A slotted device, called a finger locator, is calibrated in millimeters and is used to measure the distance from the end of the finger to the point in question. This device can be seen in figure 3, which also shows a typical radiograph of a finger.

The overall absorption coefficient, μ_B, of the bone is different from the absorption coefficient of the medullary and cortical bones. The expression which permits the evaluation of the absorption coefficients of medullary and cortical bone separately is

$$I=I_o e^{-\mu_{MB} m} e^{-\mu_{CB}(b-m)} e^{-\mu_T(t-b)} \qquad (3)$$

where

μ_{MB} is the absorption coefficient of medullary bone in cm^{-1},

μ_{CB} is the absorption coefficient of cortical bone in cm^{-1}, and

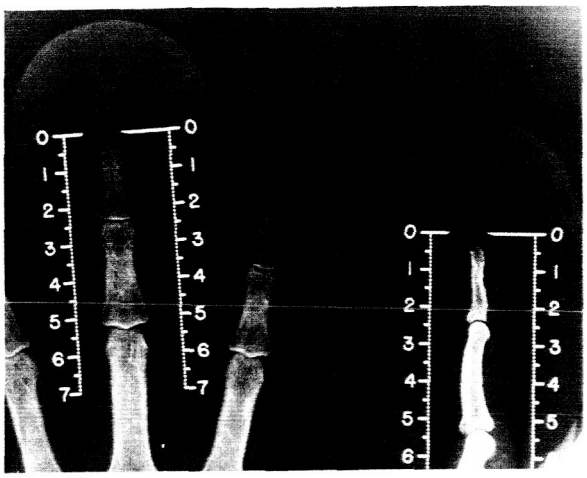

FIGURE 3.—Film of finger in locater, calibrated in millimeters. Nonscreen film with target-film distance 100 cm.

m is the thickness of the medullary bone in cm.

The other terms are defined as above. The thickness of medullary bone is found by means of the radiograph. If a value of μ_{MB} is determined separately, the absorption coefficient of cortical bone can be determined.

Bone consists of supporting tissue plus hydroxyapatite (effective atomic number 16.65). If we take a layer of hydroxyapatite, or material having the same effective atomic number and of a thickness as to give the same attenuation in the I^{125} beam as is given by an unknown bone *in vivo*, the density of the *in vivo* bone can be determined. This assumes that the mass absorption coefficient, μ_H/ρ_H, equals that of the bone, μ_B/ρ_B, and that there is negligible absorption of the radiation in the supporting tissue.

INSTRUMENTATION

The I^{125} is taken up from a solution by a small amount of ion-exchange resin which is then used as the source. This iodine containing resin is placed in a small, cylindrically-shaped plastic capsule, and is sealed by means of an "O" ring and a thin plastic cover plate. The cylinder is placed in a brass source shield with a sliding shutter which is controlled by an electromagnet, as shown in figure 4. The beam, as it leaves the source shield, is collimated by an insert of brass. Beam diameters from 0.3 to 2.0 mm have been used.

The object to be measured, i.e., the finger, is held immobile by means of two plastic plates. The plates are milled out in the region of measurement but are quite thick elsewhere. These plates are shown in figure 5.

The beam is detected by a thallium-activated, 2-mm-thick sodium iodide crystal. To prevent light and moisture from entering the crystal, it is covered on the entrance side of a 0.13-mm berryllium foil. This foil absorbs only about 1% of the radiation because of its small thickness and low atomic number. Since the photon energies I^{125} are quite low, the crystal is a very efficient detector from which very little energy escapes. The crystal is surrounded by another shield to reduce the background radiation which reaches the crystal. This shield contains a collimator hole 3 mm in diameter (see fig. 4).

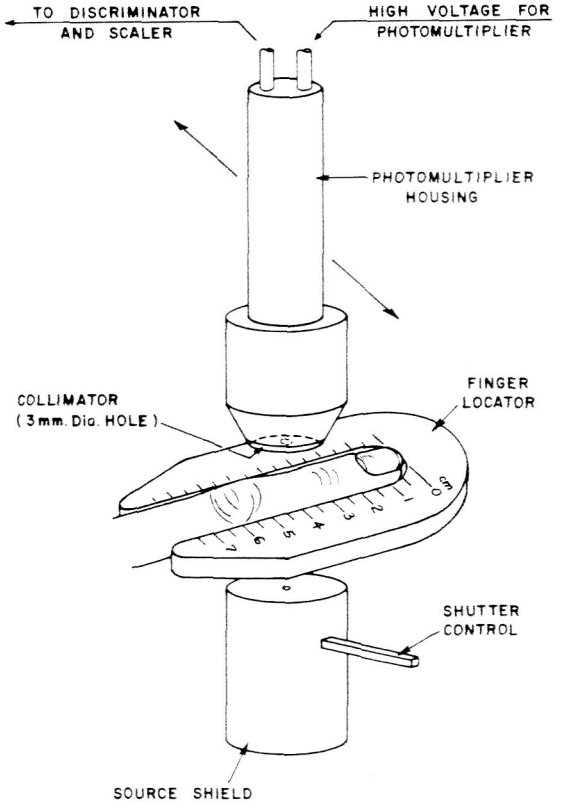

FIGURE 4.—Diagram of source and detector. The source and detector sector across immobile object.

FIGURE 5.—Instrument in use. Parallel plates hold finger immobile.

The light pulses, which result when the X- and gamma-rays interact with the crystal, are picked up by a photomultiplier. The photomultiplier is followed by a preamplifier, amplifier, and by a single channel analyzer which actually operates as a discriminator. All pulses below a preset pulse-height level are rejected. All pulses above that level are sent to a scaling circuit to be counted. A differential pulse height spectrum is shown in figure 6. A Baird-Atomic Cambridge Series Model SC107 scaler and Cambridge Model SC905 timer are used. The unit can be set to count for a present time, or a preset number of counts. The apparatus is shown in figure 5. To facilitate data recording, the total counts, if the counting time is preselected, or time, if the total counts are preselected, is automatically recorded on paper tape from a Hewlett Packard Digital Recorder. The power supply for the photomultiplier is a John Fluke Model 405.

The entire unit is normally programmed to make transmission measurements across the entire width of the finger automatically. The finger is held immobile, but the source and detector are mounted on a frame which is driven laterally across the finger by means of a drive motor coupled through an electric clutch and brake to a lead screw. Measurements are made at 1-mm intervals. During each measurement period, the source and detector are held fixed. The electric brake is energized to insure that the lead screw is not rotated, thus preventing the

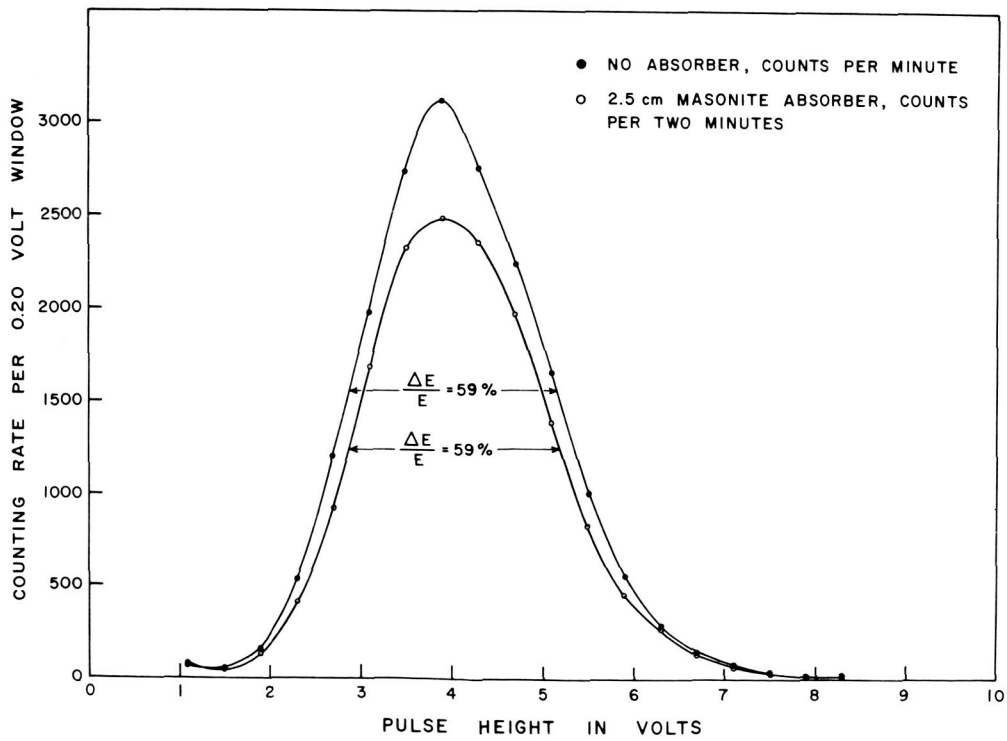

FIGURE 6.—Pulse height vs. counting rate, I^{125}.

source and detector from moving. Following a counting interval, the brake is de-energized and the clutch is energized, allowing the motor to turn the lead screw one revolution. One revolution corresponds to a 1-mm movement of the source-detector carriage. During this off period, the digital recorder is activated and prints out the time or the counts for the previous interval. Upon completion of the 1-mm movement, the measurement mode again takes over. This process is continued until the entire finger has been measured.

DATA HANDLING

Equation (2) relates the initial intensity of the beam to the intensity as altered by the presence of the finger. Solving this equation for the absorption coefficient, μ_B, gives

$$\mu_B = \frac{\ln I/I_o + \mu_T(t-b)}{b} \qquad (4)$$

where the terms of the equation are as defined above. The right-hand side of the equation contains five independently observed quantities. It is useful to consider the measurement uncertainties of each of these quantities and to establish their relationship to the uncertainty of the absorption coefficient of bone. The fundamental formula of the propagation of errors can be used:

$$\sigma_f = \left[\left(\frac{\alpha f}{\alpha x_1}\right)^2 \sigma_1^2 + \left(\frac{\alpha f}{\alpha x_2}\right)^2 + \ldots + \left(\frac{\alpha f}{\alpha x_n}\right)^2 \sigma_n^2\right]^{1/2} \qquad (5)$$

where σ_f is the standard deviation of a function f of the independently observed quantities x_1, $x_2 \ldots, x_n$ with their respective standard deviations $\sigma_1, \sigma_2, \ldots, \sigma_n$.

Considering the dependent variable to be μ_B from equation (4), one obtains

$$\sigma_{\mu_B} = \left[\left(\frac{1}{bf}\right)^2 \sigma_1^2 + \left(-\frac{1}{b1_o}\right)^2 \sigma_{1_0}^2 + \left(\frac{t-b}{b}\right)^2 \sigma_{\mu_T}^2 + \left(\frac{\mu_T}{b}\right)^2 \sigma_{t-b}^2 + \left(-\frac{\ln I/I_o + \mu_T(t-b)}{b^2}\right)^2 \sigma_b^2\right]^{1/2} \qquad (6)$$

where σ_1, σ_{1_o}, σ_{μ_T}, σ_{t-b}, and σ_b are the individual standard diviations of the quantities 1, I_o, μ_T, $(t-b)$, and b.

If a series of measurements of the above quantities is made on an individual and the standard deviations are calculated, a statement can be made about the standard deviation of computed values of μ_B from the mean value, using equation (6). It is necessary to obtain this standard deviation to be able to distinguish between the uncertainty of an individual determination and a spread of values in a population of persons. We find that it is possible to obtain values of $\sigma\mu_B$ to about $\pm 1\%$ when each of the individual quantities is carefully measured. In the quantities I and I_0, a sufficiently high number of counts must be taken and corrected for background counts.

USE OF APPARATUS

Our measurements are expressed as linear coefficients of absorption of total bone (μ_{TB}). The total thickness of the phalanx is easily measured. A bone (fig. 7) of equal total diameter but with a thinner cortex will give a lower linear coefficient of absorption. Osteomalacia bone would also give a lower linear coefficient of absorption.

We have performed a study on postmenopausal females with and without hormone therapy. We will present the results tomorrow. We have followed osteoporotic patients, male and female, on hormone therapy and have started a project to do serial measurements of patients on cortisone therapy. The apparatus can also be utilized for studies of animals, such as the rabbit. Figure 8 demonstrates the frontal and lateral views of the tibia of a rabbit. Figures 9 and 10 demonstrate the frontal and from-above views of the rabbit holder. Figure 11 shows the rabbit in place with the tibia in position to be measured.

ACKNOWLEDGMENTS

The authors would like to express their thanks to Donald Charleston, Robert Beck, and John C. Wood for assistance with the electronics problems. Dr. Paul V. Harper kindly loaned us our first I^{125} source during preliminary testing. E. Duthorn and A. Cox helped with the patient measurements.

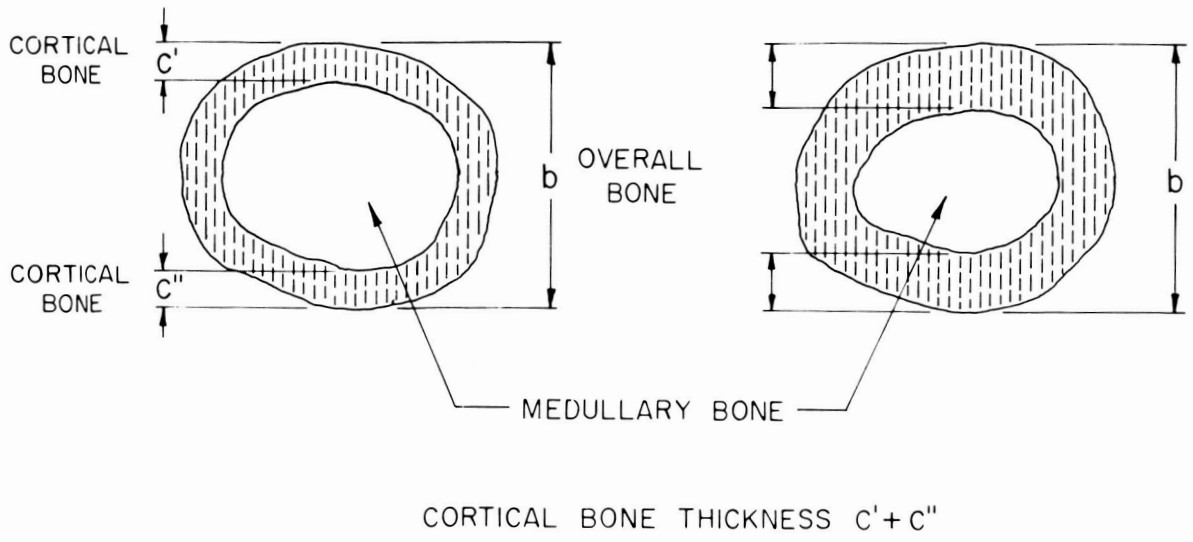

FIGURE 7.—Difference in thickness of cortical bone of normal and osteoporotic bone.

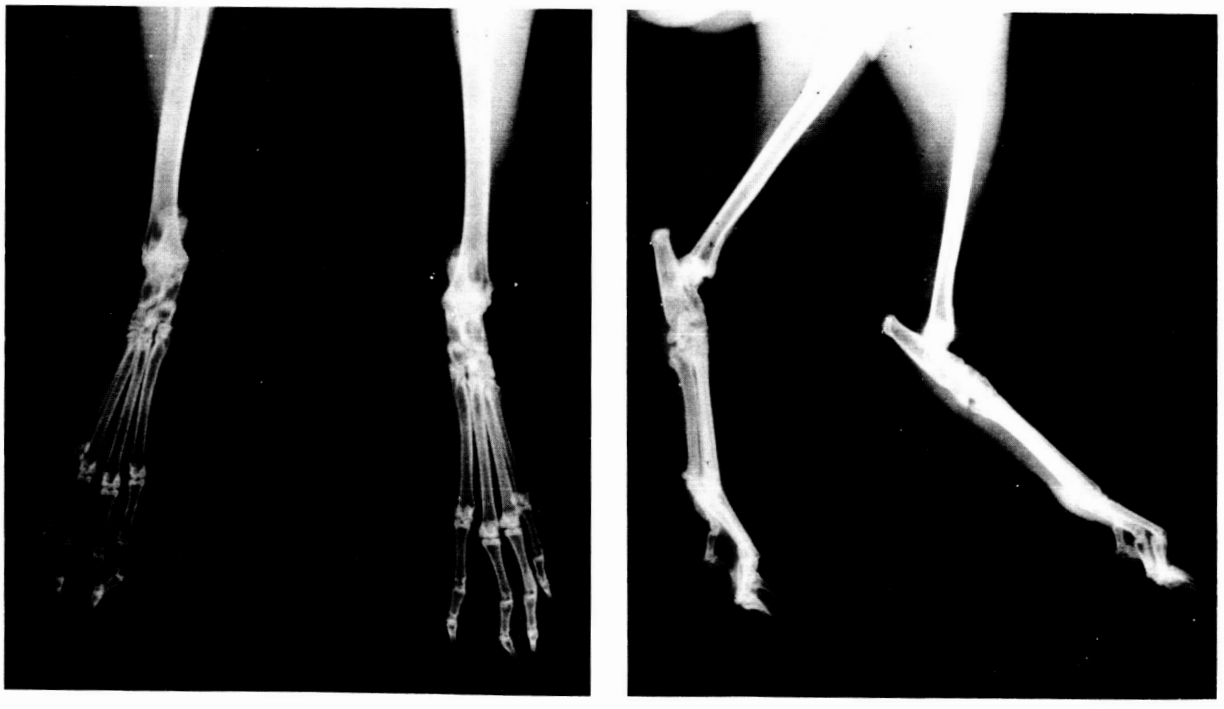

FIGURE 8.—Radiographs of rabbit tibiae in frontal (left) and lateral (right) views.

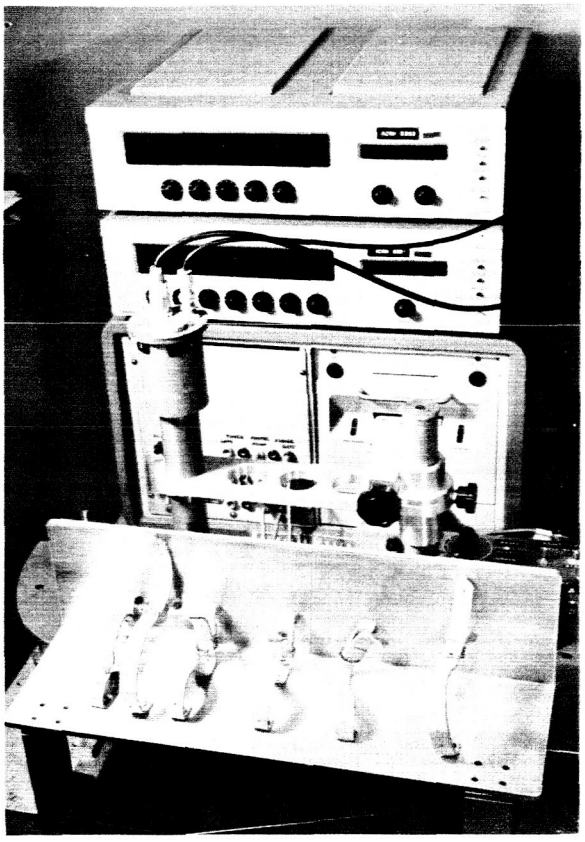

FIGURE 9.—View of rabbit holder from front.

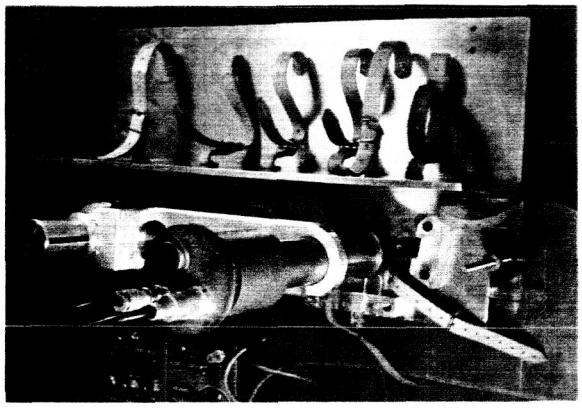

FIGURE 10.—View of rabbit holder from above.

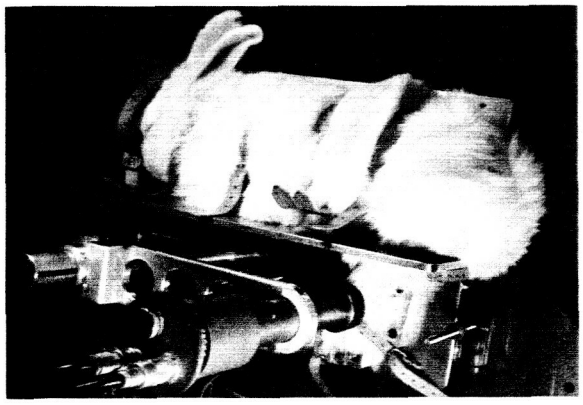

FIGURE 11.—Holder with rabbit in place to measure transmission through right tibia.

REFERENCES

ALBRIGHT, F.; and REIFENSTEIN, E. C., Jr.: Parathyroid Glands and Metabolic Bone Disease. The Williams and Wilkins Company, Baltimore, 1948.

ALBRIGHT, F.; SMITH, P. H.; and RICHARDSON, A. M.: Postmenopausal Osteoporosis, Its Clinical Features. J. Am. Med. Assoc., vol. 116, May 1941, pp. 2465–2474.

BARNETT, E.; and NORDIN, B. E. C.: The Clinical and Radiological Problem of Thin Bones. Brit. J. Radiol., vol. 34, Nov. 1961, pp. 683–692.

BOWE, J. C.; and AXEL, P.: Decay of Te^{124m}. Phys. Rev., vol. 85, Mar. 1952, p. 858.

CAMERON, J. R.; and SORENSON, J.: Measurement of Bone Mineral in Vivo: An Improved Method. Science, vol. 142, no. 3589, Oct. 1963, p. 230.

DOYLE, F.H.: Ulnar Bone Mineral Concentration in Metabolic Bone Diseases. Brit. J. Radiol., vol. 34, Nov. 1961, pp. 698–712.

GARN, S. M.; ROHMANN, C. G.; and NOLAN, P. Jr.: Studies on the Development of Compact Bone, in Normal Individuals and in Endocrine and Nutritional Abnormalities. Final Progress Report Part 1, Department of Growth and Genetics, Fels Research Institute, Yellow Springs, Ohio, Dec. 31, 1963.

GERSHON-COHEN, J.; CHERRY, N. H.; and BOEHNKE, M.: Bone Density Studies With a Gamma Gage. Radiation Res., vol. 8, June 1958, pp. 509–515.

792-955 O—66——9

HARRISON, M.; FRASER, R.; and MULLEN, B.: Calcium Metabolism in Osteoporosis. Lancet, vol. 1, May 1965, pp. 1015–1019.

HINE and BROWNELL, eds.: Radiation Dosimetry. Academic Press, Inc., New York, 1956, 90 pp.

JACKSON, H.: Problems in the Measurement of Bone Density. Brit. J. Radiol., vol. 24, Nov. 1951, pp. 613–616.

LANZL, L. H.; and STRANDJORD, N. M.: Radioisotopic Device for Measuring Bone Mineral. Symposium on Low Energy X and Gamma Sources and Applications, Illinois Institute of Technology Research Institute, Chicago, Illinois, Oct. 1964.

MAYER, E. H.; TROSTLE, H. G.; ACKERMAN, E.; SCHRAER, H.; and SITTLER, O. D.: A Scintillation Counter Technique for the X-Ray Determination of Bone Mineral Content. Radiation Res., vol. 13, July 1960, pp. 156–167.

MAYO, K. M.: Quantitative Measurement of Bone Mineral Content in Normal Adult Bone. Brit. J. Radiol., vol. 34, Nov. 1961, pp. 693–698.

NORDIN, B. E. C.: Calcium Balance and Calcium Requirement in Spinal Osteoporosis. Am. J. Clin. Nutr., vol. 10, May 1962, pp. 384–390.

NORDIN, B. E. C.: The Pathogenesis of Osteoporosis. Lancet, vol. 1, May 1961, pp. 1011–1015.

NORDIN, B. E. C.; BARNET, E.; MACGREGOR, J.; and NISBIT, J.: Lumbar Spine Densitometry. Brit. Mal. J., vol. 1, June 1962, pp. 1793–1796.

OMNELL, KARL-AKE: Quantitative Roentgenologic Studies on Changes in Mineral Content of Bone in vivo. Acta Radiol. Suppl. 148, 1957, pp. 7–86.

PUGH, D.G.: Roentgenologic Diagnosis of Diseases of Bone. The Williams and Wilkins Company, Baltimore, 408 pp., 1954.

URIST, M. R.: The Etiology of Osteoporosis. J. Am. Med. Assoc., Feb. 1959.

URIST, M. R.: The Problem of Osteoporosis. Clin. Res., vol. 6, Nov. 1958, pp. 377–385.

VIRTAMA, P.: Variations in the Ash Contents of Bones of the Extremities. Ann. Med. Exptl. Biol. Fenniae. (Helsinki), vol. 38, 1960, pp. 127–132.

VIRTAMA, P.; KAJANOJA, P.; HOPSU, V. K.; and TELKKA, A.: Density of Human Carpal, Metacarpal and Digital Bones. Ann. Med. Exptl. Biol. Fenniae (Helsinki), vol. 38, 1960, pp. 467–471.

VIRTAMA, P.; and MAHONEN, H.: Thickness of the Cortical Layer as an Estimate of Mineral Content of Human Finger Bones. Brit. J. Radiol., vol. 33, Jan. 1960, pp. 60–62.

VOSE, G. P.: X-Ray Transmission Factor in Estimating Bone Density. Radiology, vol. 71, July 1958, pp. 96–101.

COMMENTS

Dr. SMITH. I am not sure why you distinguished between compact bone and trabecular bone in the middle phalanx of the third finger. What difference does it make, because it seems to me that this is a bone whose compact portion is not too well defined at times.

Dr. STRANDJORD. It is very well defined.

Dr. SMITH. What difference does it make if it is compact?

Dr. STRANDJORD. We are counting it, too. If you did want to count cortical bone thickness, you would have to take the medullary cavity out of it. Our figures will be in linear coefficients of absorption of total bone.

Dr. URIST. This issue of the kind of bone raises the question, what is the effect of the packing of the apatite crystal, what is the relationship between the packing of the apatite crystal (the crystal orientation) with respect to the organic material in the bone and the radio density as it is measured by all of these different methods? This question always comes up when you consider that radio density is so very different in the various kinds of bone structure that you can see this immediately with the naked eye. What you see is that the least dense bone is trabecular bone. The next less dense bone is dentine. Then comes lamellar bone. The lamellar bone is less dense than the Haversian bone, which is less dense bone than the intermedullary bone of the laying hen. This bone is less dense than calcified cartilage, and cartilage is less dense than enamel.

In this bone the crystals are not oriented with respect to the fiber structure. In the calcified cartilage you have flowers of mineral. In enamel, of course, you have the beautiful large crystal structure in a chamber, always relatively oriented with respect to the long axis of the chamber. In Haversian bone it is like plywood; it goes in all directions. The question I would like to ask physicists, Dr. Cameron and

Dr. Strandjord, is what are we seeing? Are we seeing the calcium? Are we seeing the apatite? Are we seeing the relationship between the inorganic solids and the organic material? In all of these different forms of tissue are there different ground rules for each one because of, first, the kinds of crystals and crystal size, second, crystal orientation, and third, the ratio of matrix to mineral.

Dr. LANZL. I might try to answer part of that briefly, namely, that what you are seeing is absorption by the various atomic constituents. We do not distinguish whether it is in the form of a crystal or not. There is a kind of artifact, though, when you use a broad beam as against a narrow beam. The former is sensitive to positioning; the latter is not.

Dr. BABCOCK. With regard to Dr. Urist's question as to what we measure, of the measurement that we make of bone segment cut up after dissection, of the operational bone mass unit, 93% is bone ash, that is, mineral content that is left after ashing at 600 degrees. Of the remaining 7%, 3% of our measurement is fat and 4% is water and protein. We do not know how much of this 93% ash is calcium.

Dr. URIST. Let us add three more parameters. Trabecular bone is more dense than muscle. Muscle is more dense than water, and fat is less dense than water. The question is: Can these methods distinguish between organic substances and inorganic solids?

Dr. STRANDJORD. The reason we have difficulty in detecting these is that we are looking at the thickness of bone and its linear coefficient absorption, and we are lumping all of this together. For a given outside diameter of this bone, we can measure quite accurately the linear coefficient absorption. After subtracting the known soft tissue, which is measured, we can take out that factor. Then we are saying that this bone absorbs this much X-ray and is it different from patient to patient, is it different between patients with various diseases?

Dr. NORDIN. How many microcuries would you and Dr. Cameron use in order to get a continuous recording instead of an individual one in the phalanx and the forearm?

Dr. LANZL. I do not have an answer, but we prefer to do it in a fixed rather than in a moving situation. In a sense we do not use continuous recording.

Dr. NORDIN. Why do you prefer it fixed? It is much better to do it continuously.

Dr. LANZL. You run into time resolving problems of your equipment.

Dr. MALETSKOS. In one case you use a ratemeter, in the other case you use a scaler. For what they want to do, the accuracy they are getting is apparently sufficient for them. If you want to do something else, as Dr. Cameron described today, I am sure the source strength goes up quite a bit.

Dr. BABCOCK. The drive continues across the radius, using a 0.3-second time constant. To accomplish this in about 10 or 20 seconds requires a source of approximately 30 mc. To do it intermittently or to do it in a small chicken bone, driving across more slowly, requires a source of only 3 to 5 mc.

Dr. NORDIN. This compares with a dose of 15 μc being used in the other procedures.

Dr. LANZL. The figure is closer to 7.

Dr. NORDIN. In fact, there is a difference in the order of a thousand-fold.

Dr. STRANDJORD. We have used a continuous recording, too, but it is not as accurate.

Dr. MALETSKOS. It is essentially the product of time and activity that you are interested in.

Dr. CAMERON. Are your absorption data for photoelectric only, or is this just an average term including photoelectric and compton, or don't you want to get involved in that aspect?

Dr. COHEN. Isn't the first term compton?

Dr. CAMERON. The first term he said was scatter. That could be compton.

Dr. LANZL. It is scatter. I would like to say a word about "good" and "poor" geometry. If you have a beam impinging on a target of some sort and if you have a detector in so-called poor geometry, you see an attenuated beam because the primary beam is attenuated. If the beam is striking the whole absorber, you have the probability of getting scattered photons at the detector position. In this case you will get a higher reading than in the second case where you have a collimated beam, an absorber, and a detector. You have attenuation from the same target, but the intensity reaching the point will be less because you are not irradiating all of the absorber. This is one of the difficulties of broad beam or poor geometry.

Using a bone wedge nearby, and a film detector which does not have any buckeye diaphragm, the X-ray beam gives both the primary signal and a scattered signal from some other part. That introduces a complication which you do not have in the good geometry situation.

Dr. CAMERON. You use the symbol B, which is your bone thickness in various positions. How do you measure your bone thickness?

Dr. LANZL. We have a holder in which we put the hand of the individual. We take two exposures. We measure the thickness across the bone.

Dr. CAMERON. Is this minus the medullary canal?

Dr. LANZL. I include the medullary canal. In other words, I am saying that this is bone. My colleague tells me there are two different kinds of bone, but this is bone nonetheless.

We have to be careful because if the hand is not against film, there is a very slight enlarging.

So far, we have just measured this distance on the film with a lens and a reticle. This is perhaps a weak point because I think our technique might not be as good as others presented earlier today or yesterday.

Dr. MALETSKOS. I would like to return now briefly to the problem of linearization. The question that was asked yesterday was whether the machine used by Dr. Mack was designed and works in a way that makes the

transformation in such a way that the answer is in terms equivalent either to aluminum thickness or mass or bone mass. We are concerned only with the principle.

Now the question is, does the instrument do this? I am not going into the details of the instrument; I am going into what I think is the very heart of the equipment.

The instrument has a potentiometer. There is an arm which can be moved around. The signal comes in at a point, in effect, and moves the arm around to indicate a certain response. Eventually this is recorded on the paper.

Mechanically tied in with this is another potentiometer which also has an arm that moves at the same angle as the first because they are tied together, and from this another signal is tapped off. In the ordinary potentiometer, the voltage would be proportional to the distance traveled.

What you would like to do is to get a signal on the second potentiometer which is not *directly* mechanically coupled to the first. Rather, you impose a potential distribution from zero resistance to maximum resistance which is exactly equal to the shape of the calibration curve. Once you have done that, if a signal comes in which is a response and moves the first pointer so far on the potentiometer, the second automatically moves and automatically makes the analogy of going from point to point. Once you have done that you have transformed your signal into terms of aluminum thickness. Now you can do whatever you want with the result. In the original instrumentation, the final curve only was shown. Dr. Mack's equipment has another chart which shows another curve. What is wanted is the area under this curve. They happen to have an integrated unit, but that is not necessary to the argument at the moment.

The analogy is performed by mechanical coupling between these two potentiometers, one of which is adjusted by all the knobs you have seen on the diagrams so that the distribution of voltage is exactly proportional to the shape of the curve.

In my opinion, the instrument does what was intended, namely, it makes an analog transformation. The difficulty, I believe comes in the use of the word linearization. If now you want to check your instrument, you would rerun the curve through and the result would be a straight line. All this means is that you have set the distribution of potential in exactly the same shape. I think the use of the word linearization has perhaps been misleading. What you are actually doing is performing an analog transformation.

Dr. GARN. I think this probably puzzles some people. There is a point or set of points at either end of any sensimetric or densimetric curve beyond which you may not go. If you go beyond those points, further change in transmission or density (whatever way you are working it), you will not get a meaningful picture of change in wedge equivalent thickness. The important point is that perhaps this is, in effect, a danger signal. It is a point or a set of points beyond which they ordinarily will not go.

Dr. LANZL. What you are saying, if I may summarize, is that you can misuse an instrument. If you misuse it then you had better be careful what you do with the data. That is what you say when you are going to two extremes.

Dr. CAMERON. I don't think it is wrong with those two extremes. It is less accurate. It is important not to say it is wrong because it is not wrong. It can be less sensitive and this may be a problem.

Dr. GARN. It is not just a loss in sensitivity but actually very wrong and can be obtained at the two points which I have called roll-off.

Dr. ROCKOFF. I now understand the operation that Dr. Mack was attempting in theory. I would like to explain why this problem came up. Among the data that were sent to us to evaluate was one of Dr. Mack's publications which showed the nonlinearized original film response curve (what she refers to as the linearized tracing on the same scale). No scale values were given and nowhere in the text was there a reference to different scales. Believing they were put on the same scale added some confusion to our analyses. It is now clear that the principle of the instrumentation is valid and, if the work was being done correctly, that this transformation could be done in a valid fashion.

The questions left to be answered, which are not within the domain of this conference *per se*, concern the precision and accuracy and inherent biases in the system that is being used. I do not believe that the linearization technique, as Dr. Mack calls it, if it is being used correctly, is a source of that bias at the moment.

Dr. JENKINS. I would like to thank Dr. Rockoff and Dr. Zelen for assisting NASA and this working conference by examining Dr. Mack's technique. They have also recommended some very critical experiments which Dr. Mack has been conducting recently, which she presented as an important part of her paper.

I have participated in many of the detailed discussions and I think that some of the homework that was done for this working conference has pointed out a number of the critical problems which this working conference could discuss. If this had not been done, we would not have made as much progress.

Dr. MALETSKOS. We have been discussing different kinds of electromagnetic radiation and the use of densitometry this morning. There are other methods of getting different kinds of electromagnetic radiation.

The next paper will explain one method which can eventually have use in this densitometric technique.

Design and Applications of β-Excited X-ray Sources

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T. G. Stinchcomb
IIT Research Institute

Although the outputs of isotopic X-ray sources are not as high as those of conventional machine sources, they do have many valuable features such as portability, compactness, stability, low initial and maintenance cost, and no power requirement. The prospective uses of these sources is therefore quite promising.

There are three classes of isotopic X-ray sources: direct X-ray or γ-emitters, β-excited X-ray sources, and α-excited sources. The direct X-ray or γ-ray is either emitted from the nucleus following electron capture or β-decay or is emitted from the atomic K and L shells following electron capture or internal conversion. I^{125} and Am^{241} are examples of this type of isotopic source and have been discussed in previous papers. In the β-excited X-ray source the X-rays are produced by the stopping of β-rays much as the X-rays are produced in a conventional X-ray machine by the stopping of electrons. The continuous bremsstrahlung spectrum produced by the stopping of the betas is peaked by the K-edge in self-absorption of the X-rays in the target. In addition there is K shell X-ray emission both from atoms ionized directly by the betas and from atoms ionized by the bremsstrahlung (fluorescence). The α-excited sources emit X-rays only by K shell X-ray emission from atoms ionized directly by the alphas. Because there are no bremsstrahlung X-rays, the intensity from α-excited sources is relatively low; but this disadvantage is offset by the extreme sharpness of the K X-ray line from the target.

Because the β-excited X-ray sources offer much flexibility in choice of geometry and source and target materials, the development and application of β-excited X-ray sources have been carried out initermittently over the past 14 years at IIT Research Institute and elsewhere. This paper summarizes the research performed at IITRI by many investigators, the more extensive work having been done by L. Reiffel, C. A. Stone, L. Voyvodic, and Italo Filosofo. The investigations consisted of four phases: first, an experimental survey on X-ray production using sources of several energies, targets of various thicknesses and atomic numbers, and various geometries; second, an analytical study to understand and interpret these experiments; next, an application of the results of the study to design and build several high level intensity prototype sources having characteristics optimized for given applications; and finally, the utilization of the prototype sources in several typical applications to demonstrate the widespread potential uses of these sources.

Some of these investigations were sponsored by the Division of Isotopes Development of the U.S. Atomic Energy Commission.

EXPERIMENTAL SURVEY OF X-RAY PRODUCTION

Because of their ready availability, low cost, and long half-lives, our experimental work has been concerned mainly with the use of the fission-product β-emitters: Pm^{147}, Kr^{85}, and

Sr-Y^{90}. These emitters are typical of low, medium, and high energy sources, respectively. Other workers have used tritium (absorbed in titanium or zirconium), Ca^{45} and P^{32}.

An extensive experimental survey has been made of the X-ray output from sources using these β-emitters with a variety of target materials ranging from copper to uranium. Five different geometrical arrangements have been used:

1. Transmission, where the target material is placed between the β-source and the X-ray detector;
2. Reflection at 180° where the β-source is placed between the target and the detector;
3. Sandwich, the target material on both sides of the beta source;
4. Reflection at 90°, where the X-rays are detected at 90° to the β-particle direction, the target being at 45° to both; and
5. Mixture geometry, with the β-emitter intimately mixed with the target material.

A β stopper of lucite was inserted between the β-source and the detector in all those measurements where the target thickness was not enough to stop all the betas. The X-ray spectra and intensities were studied primarily with a NaI(Tl) scintillation spectrometer and a pulse height analyzer. In all the measurements the detectors have been shielded, and the X-rays detected through a lead collimator lined with cadmium and copper layers to eliminate high energy fluorescent lead X-rays.

The detailed results of these studies have been reported quantitatively and are in approximate agreement with similar results by other investigators as reviewed by starfelt et al. They may be summarized qualitatively by the following general statements:

1. The sandwich geometry is shown to be the most efficient geometry for high energy β-emitters (such as Sr-Y^{90}).
2. The mixture geometry is shown to be the most efficient geometry for low energy β emitters (such as Pm^{147}).
3. The 90° geometry shows the best peak purity (i.e., the least intensity at energies different from the K X-ray peak).
4. For sandwich and transmission geometries, the intensities of the K X-ray peak first increase and then decrease as the target thickness is increased, the optimum thickness being defined as that at which the maximum occurs in the intensity of the K X-ray peak.
5. For all geometries, an increase in target thickness is accompanied by an increased attenuation of the bremsstrahlung continuum at energies just higher than the K absorption edge which results in an increased narrowing of the peak.
6. Optimum thicknesses are lower for lower β energy.
7. The peak intensity for a given target material (i.e., a given K peak energy) increases with increasing β energy.
8. The peak purity for a given target material (i.e., a given K peak energy,) generally decreases (due to an increasing bremsstrahlung high energy tail) as the β energy increases beyond values an order of magnitude higher than the K peak energy.

ANALYTICAL STUDY OF X-RAY PRODUCTION

To aid in the design of new sources with specific performance characteristics, an approximate mathematical formulation has been developed by analytically studying the results of the experimental survey just described. One can assume an isotropic distribution of the X-rays at their production by β excitation and can consider the propagation of these X-rays in a limited solid angle about a direction normal to external slab targets. Viewing the X-ray yield as governed by the two processes, production by β excitation and self-absorption in these slab targets, the yield $Y(W,t)$ of photons of energy W per β particle striking a target of thickness t has been approximated by the expression

$$Y(W,t) \approx N(W) \int_0^t A_x(W,t') A_\beta(W,t') dt'$$

where $N(W)$ represents the total number of X-rays produced per beta stopping in a thick target, $A_\beta(W,t')dt'$ represents the fraction of these produced in the differential thickness dt', and $A_x(W,t')$ represents the absorption function for the X-rays from the point of their production to their emergence from the target.

The absorption of betas in the external target can be represented by an exponential law (Evans, 1955). For the β absorption coefficient we have used $\mu_B=8/R_{max}$ which reflects the experimental evidence of a β half thickness of about one-tenth the maximum range. We have further assumed that the fraction of the X-rays produced in a differential target thickness dt' is equal to the fraction of the betas absorbed in the same thickness.

For the source-target mixture we have assumed that all the betas are absorbed near the point of their emission, and hence $A_B\,dt' \approx dt'/t$, i.e., equal probability of X-ray emission throughout the target thickness. This is a good approximation in the case of low energy β emitters for which this geometry is particularly attractive.

With these functions the photon yield functions for β-excited X-ray sources are obtained as follows:

Transmission Geometry

$$Y_T(W,t)=N(W)\,\frac{\mu_\beta}{\mu_\beta-\mu_x}\,(e^{-\mu_x t}-e^{-\mu_\beta t})\ \text{photons}/\beta \tag{1}$$

$$t_{opt}=\frac{\lg\mu_x-\lg\mu_\beta}{\mu_x-\mu_\beta} \tag{2}$$

Reflection Geometry

$$Y_R(W,t)=N(W)\,\frac{\mu_\beta}{\mu_\beta+\mu_x}\,[1-e^{-(\mu_\beta+\mu_x)t}]\ \text{photons}/\beta \tag{3}$$

Sandwich Geometry (Back Target, $t>R_{max}$)

$$Y_S(W,t)=\frac{N(W)}{2}\left[\frac{\mu_\beta}{\mu_\beta-\mu_x}\,(e^{-\mu_x t}-e^{-\mu_\beta t})\right]+\frac{\mu_\beta}{\mu_\beta+\mu_x}\,e^{-\mu_x t}\ \text{photons}/\beta \tag{4}$$

$$t_{opt}=\frac{\lg(\mu_\beta+\mu_x)-\lg 2\mu_x}{\mu_\beta-\mu_x} \tag{5}$$

Source-Target Mixture (Low Energy Emitters)

$$Y_M(W,t)=N(W)\,\frac{(1-e^{-\mu_x t})}{\mu_x t}\ \text{photons}/\beta\ (t\geq R_{max}) \tag{6}$$

Calculated optimum thicknesses based on these results are compared with experimental optimum thicknesses in table I. The agreement is encouraging, considering the approximate nature of the treatment.

For the thick target production functions $N(W)$, approximate expressions were used for the bremsstrahlung continuum and for the characteristic radiation due to K-shell ionization by the β-particles. These expressions were adapted from Evans, Heitler and Burhop. Table II lists the resultant formulas along with numerical estimates for tin and lead targets. In this table, N_{bp} denotes the contributions to the K peak from the portion of the continuum (between W_1 and W_2) which leads to bremsstrahlung peaking just below the K absorption edge; N_{bt} represents the integral photon production at energies above the absorption edge. This term is helpful for estimating the degree of K peak purity and also for the contribution of fluorescent K X-rays due to photoelectric absorption within the target. The quantity N_{ks} is the X-ray production due to K shell ionization. The total number of photons produced in the K peak per β-particle absorbed in a thick target is then given by

$$N(W)=N_{bp}+N_{ks}+GN_{bt}$$

G represents the fraction of the photons in the high energy bremsstrahlung tail which, upon absorption, give rise to a fluorescent K photon. This factor depends on the geometry. The dependence is such as to suggest a low value of G for transmission, sandwich, and reflection geometries, and a high value for 90° geometry and mixture geometry. In table II, two values are shown for the calculated yield $Y(W,t_{opt})$: one computed for $G=O$ and one for $G=1$. Also shown are the measured values from the results of the experimental survey. These values, in general, lie below the calculated values by factors of 2 or less. The agreement is better for the higher energies, which may be

TABLE I.—*Calculated and Experimental Optimum Thickness of Slab Targets for Tin and Lead K Photon Peaks*

β source		Pm^{147}	Kr^{85}	Y^{90}
E_{max}—MeV		0. 226	0. 67	2. 2
$T_{1/2}$—yrs		2. 6	10. 3	27. 7
R_{max}—mg/cm²		50	250	1100
$Sn(1/\mu_k=110$ mg/cm²):				
Sandwich	Exp	12	--------	70
t_{opt}—mg/cm²	Calc	15	36	58
Transmission	Exp	18	54	110
t_{opt}—mg/cm²	Calc	18	55	124
$Pb(1/\mu_k=660$ mg/cm²):				
Sandwich	Exp	------	--------	--------
t_{opt}—mg/cm²	Calc	25	79	185
Transmission	Exp	30	100	250
t_{opt}—mg/cm²	Calc	31	100	272

TABLE II.—*Calculated and Experimental Optimum X-Ray Yields From Slab Targets for Tin and Lead K Photon Peaks*

β-Source	Pm^{147}		Kr^{85}		Y^{90}	
Z of target	50(Sn)	82(Pb)	50(Sn)	82(Pb)	50(Sn)	82(Pb)
N_{bp}	0. 13	0. 14	0. 46	0. 56	1. 5	1. 9
N_{bt}	0. 36	0. 15	2. 0	1. 7	10. 7	11. 5
N_{ks}	0. 40	0. 06	2. 0	0. 2	6. 6	0. 7
$N(W)$ $G=0$	0. 53	0. 20	2. 46	0. 76	8. 1	2. 6
$N(W)$ $G=1$	0. 89	0. 35	4. 46	2. 46	18. 8	14. 1
$Y(W,t_{opt})$ sandwich $G=0$	0. 44	0. 19	1. 4	0. 65	2. 0	1. 6
$Y(W,t_{opt})$ sandwich $G=1$	0. 74	0. 33	2. 5	2. 1	4. 7	8. 8
$Y(W,t_{opt})$ transmission $G=0$	0. 45	0. 19	1. 5	0. 65	2. 7	1. 7
$Y(W,t_{opt})$ transmission $G=1$	0. 75	0. 33	2. 7	2. 1	6. 2	9. 3
$Y(W,t_{opt})$ transmission experimental	0. 3	<0.1	0. 7	1. 0	2. 2	3. 3

Formulas Used:

$$N(W)=N_{bp}+N_{ks}+GN_{bt}$$

$$N_{bp}\approx 2k\left(\frac{E_{rms}}{E_{max}}\right)^2 Z\left(\frac{E_{max}}{W}-1\right)(W_2-W_1)$$

$$N_{bt}\approx 2k\left(\frac{E_{rms}}{E_{max}}\right)^2 ZE_{max}\left(1g\,\frac{E_{max}}{W_{ab}}-1+\frac{W_{ab}}{E_{max}}\right)$$

$$N_{ks}\approx 5.4\times 10^5\left(\frac{E_{av}}{E_{max}}\right)\omega_K(Z)Z^{-4}E_{max}\text{ for }E_{max}\geq 1\text{ MeV}$$

$$N_{ks}\approx 1.5\times 10^6\omega_K(Z)Z^{-4}\left[\left(\frac{E_{rms}}{E_{max}}\right)^2 E_{max}{}^2-W_{ab}{}^2\right]\text{ for }E_{max}\leq 0.1\text{ MeV}$$

Values Used:

$$k=7\times 10^{-4}\text{MeV}^{-1};\ \frac{E_{rms}}{E_{max}}=0.45;\ \frac{E_{av}}{E_{max}}=0.33;\ \omega_K=1$$

due to less absorption of betas in emerging through the windows of the source. It is encouraging that this rough analytical treatment gives results agreeing as well as this, for one can now use this method of analysis to predict K photon yields correct to within a factor of 2.

PROTOTYPE SOURCE DESIGN

The results of the analysis decribed above furnish a basis for designing isotopic X-ray sources. Maximum output is obtained with configurations corresponding to optimum target dimensions, but limitations exist due to the finite specific activity of the β-emitter and to the desired size of the isotopic source. Using external targets, the thickness of the β-source, including encapsulation, must be small as compared with the maximum β-range. If a small X-ray focal spot is desired, the volume of the β-emmitter and the maximum X-ray output are limited. When large area X-ray sources are acceptable or desired, a limit in the X-ray output is determined only by the area of the X-ray source; and a high X-ray flux can be achieved.

As in the external target geometries, a limit in the maximum X-ray output per unit of source area also exists for the source-target mixtures. Here, however, the X-ray output is an asymptotic function of the source thickness; and considerations of the costs of β-emitter and source fabrication can indicate which fraction of the X-ray saturation output is most convenient to achieve.

In designing an isotopic X-ray source, the desired X-ray spectral distribution is the major factor in the choice of the β-emitter. The experimental and analytical results of the previous paragraphs indicate that K peak yield and "purity" depend strongly on β-energy. For X-ray outputs peaking at characteristic radiation of medium Z targets (from zirconium to tungsten), the low energy β-emitters are the most advantageous. For high Z targets (lead, uranium), only the high energy β-emitters give a high yield of X-rays. Although high energy betas produce, in general, a large amount of high energy bremsstrahlung, the peak purity is, at least for high Z targets, satisfactory for most of the source applications.

As far as K X-ray yield and purity is concerned, one should follow the general rule of high energy β-emitters combined with high Z targets and low energy betas with medium Z targets. This rule is, however, limited in applicability to the design of compact isotopic sources. When a large size X-ray source can be tolerated, there exists more flexibility in β-emitter-target combination. For example, a substantial improvement in peak purity can be obtained if a loose geometric configuration, such as the 90° reflection target, can be used.

Strontium-90 Source Design

As discussed above, we considered the Sr^{90} a very suitable β-emitter for an isotopic X-ray source using high Z target material in a sandwich geometry. Treating the specific case of an X-ray spectrum peaking at about 100 keV (uranium target), an optimum thickness of 230 mg/cm^{-2} results.

A Sr^{90} source material very suitable for this type of X-ray source design seems to be the $SrTiO_3$ manufactured in the form of very stable ceramic pellets by ORNL. For a β-source thickness equivalent to two relaxation lengths (~300 mg cm^{-2}) and with the specific activity of 40 Ci per gram of ceramic compound, the "effective" β-activity is about 12 Ci per square centimeter of source area.

The K X-ray output, evaluated for $t_{opt}=230$ mg/cm^{-2} and a β-activity of 12 Ci per square centimeter, turns out to be 10^{10} photons/sec per square centimeter of source area, or about 25 "mCi" of K photons per curie of the β-emitter. At one foot distance from a source of area 1 cm^2, the flux of K X-rays is about 10^6 K photons/sec^{-1} cm^{-2}; and the dose rate is about 200 mr/hr.

The spectral distribution of X-rays from a Sr^{90} source with a uranium target is quite similar to that obtained with a lead target. The energy distribution of X-rays exhibits a K peak of high purity, with a ratio of about 1:1 for the

K peak and the bremsstrahlung tail components. This spectral distribution and X-ray flux that can be obtained suggested that this source should be particularly suited to thickness gauging, industrial radiography, and composition analysis by backscattering technique. Its performance is expected to be quite similar to that of the 129-day γ-emitter Tm^{170}, with the advantage of a much longer half-life. The cost of the X-ray output from the Sr^{90} and high Z targets is moderate, of the order of one dollar or less per millicurie of K photons.

Promethium 147 Source Design

The most efficient geometry for Pm^{147} is the source-target mixture where the full energy of the betas is dissipated in the target material. Maximum X-ray output from Pm^{147} can, of course, be obtained when the β-emitter is not mixed with a target compound and the characteristic X-rays are produced in the promethium itself.

For Pm^{147}, the analytically calculated value of the photon production function in the energy range from 30 to 50 keV about the 38.5 keV K X-ray of promethium is:

$$N_{bp} \approx 1.6 \times 10^{-3} \text{ photon}/\beta$$

$$N_{ks} \approx 1.0 \times 10^{-3} \text{ photon}/\beta$$

$$GN_{bt} \approx 0 - 2.9 \times 10^{-3} \text{ photon}/\beta$$

(depending on source geometry).

Assuming a value for GN_{bt} intermediate between its maximum and minimum contribution, the total production function, $N(W)$, for a promethium source should be approximately 4×10^{-3} photons/β. The X-ray output from such a source will be given, therefore, by

$$\frac{AaN(W)(1-e^{\mu_x t})}{\mu_x} \text{ photon/sec}$$

where A is the activity per gram, a is the source area in square centimeters, μ_x is the K X-ray absorption coefficient in promethium (μ_x= 4.66 cm^2/g at 40 keV), and t is the source thickness in grams per square centimeter.

The prototype source of this geometry consisted of 250 curies of Pm^{147}; this is approximately 366 mg of Pm_2O_3. The oxide was compressed and sintered into a pellet approximately 7 mm in diameter by 2 mm thick. The pellet was then encapsulated in an aluminum container ½-inch in diameter by ¾-inch long, with a 15 millimeter aluminum end window. The source was then placed in a stainless steel and hevimet housing with a movable shutter, figure 1. This provides the necessary shielding for ease of handling and allows controlled exposures.

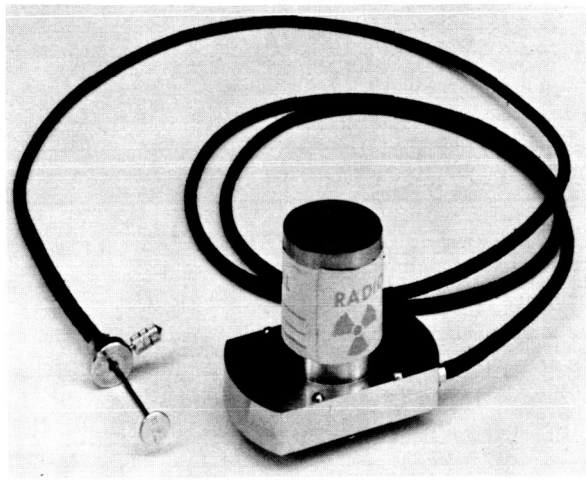

FIGURE 1.—Pm^{147} source holder.

The spectrum obtained has a pronounced peak in the region about 40 keV, due primarily to the K X-rays of promethium.

The spectral measurements show that the flux of photons with energy around 40 keV is 5.5×10^5 photons/cm^2/sec at one foot from the source, compared to the calculated flux of 8×10^5 photons/cm^2/sec at one foot. Since the 15 millimeter aluminum window absorbs ~5.5% and since ~10% of the K-shell vacancies result in emission of Auger electrons and not K X-rays, the calculated flux is reduced to 7×10^5 photon/cm^2/sec at one foot. There is, therefore, fair agreement between the actual and theoretical X-ray yield. The total flux of photons of all energies as calculated from the spectral data is $\sim 8 \times 10^5$ photon/cm^2/sec at one foot. The cost of the Pm^{147} used for the prototype source was $12.00 per curie.

UTILIZATION OF THE PROTOTYPE SOURCES IN TYPICAL APPLICATIONS

The performance of the isotopic sources discussed in the previous sections has been investigated in some typical applications. The applications which are useful in connection with bone densitometry are mass thickness gauging by X-ray attenuation and radiographic inspection.

For mass thickness of gauging, an important parameter is the spectral distribution of the X-rays since it determines both the total attenuation and the rate of change of attenuation with thickness. The statistical fluctuations depend on the former and the sensitivity on the latter. If the ideal conditions of exponential absorption are obtained, the optimum choice of X-ray energy is that for which the absorption coefficient is twice the reciprocal of the thickness to be measured. As an example, assuming bone to be pure calcium phosphate of density 3 gm/cm^3, the optimum energy for a 1-cm thickness is 47 keV. True exponential absorption is not realized in most practical situations; however, one can usually adjust the X-ray flux and the peak energy until the optimum combination of sensitivity and statistical fluctuation is achieved. β-excited sources represent a particularly attractive solution to these adjustment problems since their X-ray energy distribution can be varied according to the choice of target material and thickness.

Results of studies of mass thickness gauging which are applicable to medical technology are those made for lucite and aluminum since the average atomic numbers of tissue and bone, respectively, are similar. For aluminum (and thus for bone,) the promethium source gives the highest readability (which is defined as the ratio betwen the sensitivity and the statistical fluctuation). For lucite (and tissue) the use of a target of lower atomic number such as zirconium gives the highest readability.

In applications to a two-component system where attenuation measurements are made at two different X-ray energies one has to be very careful to insure that any pair of measured attenuations always indicate unambiguously a pair of thickness values for the two components, regardless of the arrangement of the components in the system. This is best accomplished by making the sources as monoenergetic as possible and by reducing the scattering to negligible amounts. At last October's Symposium on Low Energy X and Gamma Sources and Their Applications, Sorenson and Cameron reported the development of such a technique for a system in which the two components were fat and muscle. They used Am^{241} (60 keV) and well-filtered I^{125} (27 keV) as the two sources. They have extended this technique using the same sources to the bone-muscle system as reported earlier in today's symposium.

Also at last October's symposium, Goodman and Levin reported the development of a similar technique for measurements on the bone-muscle system in which were used fluorescent X-ray sources stimulated by a β-excited X-ray source. Dr. Levin, who is Chief Radiologist at Michael Reese Hospital, was unable to attend today's symposium because of previous commitments. The β-excited X-ray source he used was the 250 Ci Pm^{147} source. The strong peak at 38 keV is excellent for producing the 32 keV barium K X-ray in a disc of barium sulphate, and the continuum above 78 keV is fine for producing the 66 keV X-ray from a platinum foil. This technique measures bone mineral content in the lower arm with a reproducibility of 2% or better. Thus, changes in bone mineral content can easily be followed by this technique during the course of a disease or other processes causing osteoporosis. This technique has the advantages that the X-ray energies can be varied to give optimum combination of sensitivity and statistical fluctuation. The choice of barium and platinum by Goodman and Levin was not such an optimum choice but was dictated by the availability of the promethium source. For example, a β-excited source, using promethium betas to excite a uranium target, gave a spectrum with two peaks at 15 keV and 97 keV which can be used to stimulate monoenergetic K X-ray lines from a bromide compound (12 keV) and a bismuth foil (75 keV), respectively.

For radiographic inspection applications, the promethium source was tested since its parameters were chosen to give as high an intensity as possible from an approximate point source.

As examples of medical radiography, several exposures were taken using the Pm^{147} source and Kodak Royal Blue Medical X-ray film with Patterson Lightning Special Intensifying screens. This film-screen combination has a fast response to the low energy X-rays and thereby minimizes exposure times. To provide a comparison, figure 2 shows an exposure of a human skeletal hand made with a conventional hospital X-ray machine. This exposure has good diagnostic value because the definition

FIGURE 2.—Radiograph of skeletal hand, using conventional X-ray machine and film.

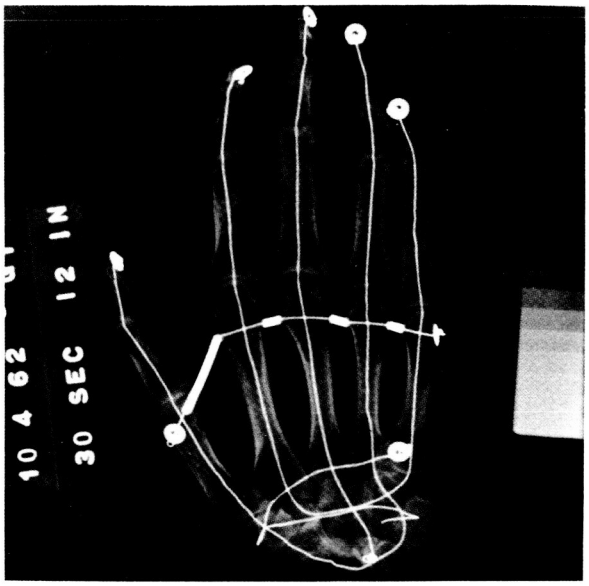

FIGURE 3.—Radiograph of skeleton hand, using Pm^{147} X-ray source and Kodak Royal Blue medical film with Patterson Lightning Special intensifying screen: 30-second exposure, 12-inch source to film distance.

and contrast of the bone fine structure is very good. An exposure of the same skeletal hand using the Pm^{147} β-excited X-ray source is shown in figure 3. The source to film distance was 12 inches, with the object placed on the film cassette, and the exposure time was 30 seconds. The bone fine structure is visible but does not have the sharpness as in the previous figure. This is probably due to penumbra effects caused by the 7-mm source size and to the graininess of the fast speed film-intensifier combination used. A similar exposure made wth a 24-inch source to film distance and a 140-second exposure time improved the quality of the fine structure only very slightly and does not appear to justify the increase of exposure time.

It may be possible to perform radiographic bone densitometry measurements with this source and film system in situations which exclude a conventional X-ray machine. We urge that radiographs using this source be evaluated for this application.

A Polaroid X-ray film unit coupled with an isotopic X-ray source represents a completely portable X-ray system. Several medical radiographs were made to evaluate the results of

FIGURE 4.—Radiograph of human hand, using Pm^{147} X-ray source with polaroid film: 40-second exposure, 9-inch source to film distance.

this system. Figure 4 shows one of these radiographs. Since some of the methods of bone densitometry require measurements of bone dimensions, it seems that a β-excited X-ray source would be able to provide these dimensions in situations which exclude a conventional X-ray machine.

SUMMARY AND CONCLUSIONS

Our investigations indicate that β-excited X-ray sources are extremely useful as complementary devices to the presently existing machines. The small size and portability of these isotopic sources make them very attractive for field uses and for nondestructive testing problems both in industry and in medicine where the situation limits the use of conventional X-ray machines. The relative monochromatic output of such sources is another advantageous feature. The only limitations of β-excited X-ray sources are centered around the limited photon outputs per unit source area.

Approximate formulae for the design and optimization of these sources have been derived, tested, and found satisfactory.

Transmission measurements of bone mineral content using a β-excited X-ray source show great promise, since monoenergetic fluorescence K X-rays can be used from almost any element. The technique of Goodman and Levin can be further developed to increase the sensitivity and the statistical accuracy.

The use of radiographs taken with β-excited X-ray sources for measurements of bone dimensions is also a possibility.

REFERENCES

BURHOP, H. L.: Proc. Camb. Phil. Soc., 36, 43, 1940.

EVANS, R. D.: The Atomic Nucleus, McGraw Hill, 1955.

FILOSOFO, I.: "Isotopic Sources of Secondary Radiation," ARF 1122–27, Armour Research Foundation, Chicago, Ill., February 1, 1961.

FILOSOFO, I.; REIFFEL, L.; STONE, C. A.; and VOYVODIC, L.: "Design and Characteristics of Beta-Excited X-Ray Source," Radioisotopes in the Physical Sciences and Industry, International Atomic Energy Agency, Vienna, 1962.

GOODMAN, L.; LEVIN, B.: "In-Vivo Measurement of Bone Material Content by Transmission Techniques," paper presented at the Symposium on Low Energy X and Gamma Sources and Applications at IIT Research Institute, Chicago, Ill., October 20–21, 1964, proceedings to be published by the AEC.

HEITLER, W.: The Quantum Theory of Radiation, Oxford University Press, 1944.

REIFFEL, L.: Nucleonics 13, No. 3, 22, 1955.

REIFFEL, L., and HUMPHREYS, R. F.: Proc. Int. Conf., Geneva, vol. 15, 291, 1956.

REIFFEL, L.: U.S. Patent 2,797,333 "X-Ray Source," June 25, 1957.

REIFFEL, L.: U.S. Patent 2,928,944 "Apparatus for X-Ray Fluorescence Analysis," March 15, 1960.

SORENSON, J. A.; and CAMERON, J. R.: "Body Composition Determination by Differential Absorption of Monochromatic X-Rays," paper presented at the Symposium on Low Energy X and Gamma Sources and Applications at IIT Research Institute, Chicago, Ill., October 20–21, 1964, proceedings to be published by the AEC.

STARFELT, N.; CEDERLUND, J., and LIDEN, K.: Jr. Appl. Rad. and Isotopes, vol. 2, 265, 1957.

COMMENTS

Dr. RICH. You seemed to indicate that Goodman and Levin spin a source so that they get two different beams alternating. Did I misunderstand that point?

Dr. STINCHCOMB. They have to take a traverse one way with a plot number. Then they switch it 180° and take a traverse the other way.

Dr. RICH. What particular advantage does use of two energies give?

Dr. STINCHCOMB. Goodman and Levin used 67 and 38 because our promethium source was available to them. Our promethium source, as you remember from the spectrum, had a big peak at about 40, so that it was just right for exciting the K spectrum, while radiation in the tail above 67 keV was high enough to excite the high energy one. Someone can go through all these equations and find out exactly what the best energies to use might be. Perhaps you have done that and found that Iodine and Americium satisfied exactly.

Dr. CAMERON. I would like to make a comment in answer to Dr. Rich's question. It is obvious that you can get measurements with only one wavelength as Dr. Strandjord and Dr. Lanzl and we have done. The one advantage of having the two wavelengths is that the thickness of the material you are measuring can be cancelled out. It enters in as a common element and you can essentially get rid of the thickness. This is not a serious problem. I do not personally think it is worthwhile having two wavelengths just to get rid of a relatively simple physical measurement.

In answer to the other question, though, about the optimum energies, this is not easy to answer in one sentence because it depends on the thickness of the material you are measuring. For example, on the thigh to go through a large amount of material, you have to use a higher energy in order to have statistical information left on the output beam. If you are going through very thin bone, you might do something on the order of 20 keV, thin beams; Cd^{109} would be a good one and we hope to investigate it. In general, any energy will do it.

Dr. STINCHCOMB. You can get any energy you want because you can make this source peak at any part of the spectrum. If you are not satisfied with the sharpness, then you can use this technique to sharpen it up further and again you can choose whatever you want.

Theoretically, one could extend this to two, three, or as many as you want, and go into calcium and phosphorus content. Whether you can get any kind of accuracy with multicomponent systems is something I certainly am not prepared to say.

Dr. CAMERON. I want to throw some cold water on it. I think it is very possible in *theory*.

Dr. STINCHCOMB. That is all we are saying.

Dr. CAMERON. There are a lot of things that are possible in theory. When you get beyond two different wavelengths, you have to spread them a long way. Putting other wavelengths in between is going to be of very marginal value.

Dr. STINCHCOMB. If you took the one that had a uranium K peak and put in fluorescence, then you could get a two-component system that could be quite widely separated. I leave it to you if this is applicable in bone densitometry.

Dr. MALETSKOS. As I indicated in the beginning, we have been talking about one physical characteristic, namely, transmission of electromagnetic radiation through material. There are many other physical principles that one can apply to the problem. Another is going to be described in the paper to be presented now.

*Sonic Measurement of Bone Mass**

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Velocity of sound varies according to the density and elastic properties (for solids) or compressibility (for liquids and gasses) of the medium through which it passes, and is greater in solid than in liquid media (Carlin, 1960). Thus, when a pulse of sound is passed through a limb which contains bone and soft issues, such as muscle, the velocity will be greater in the bone than in the soft tissues and the time necessary for sound to traverse the limb will depend upon the distance of the sonic path in each tissue. If the velocities in soft tissue are similar and sufficiently different from that in bone, and when the total distance is constant, the transit time can be used to measure the amount of bone present. Potentially, the same principle can be used to measure the proportion of any two substances (muscle and bone, liver and fat, or muscle and air) through which the sound passes, irrespective of their geometric configuration.

We have previously reported briefly on application of this principle to measure bone mass (Rich et al., 1963). In this paper we will describe the instrument by which these measurements are made and give results which point to a general applicability of this method.

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METHOD

Professor Rushmer's group at the University of Washington have developed an instrument, the "sonodistometer," which measures the time between transmission and reception when a burst of sound is passed between two piezoelectric crystal transducers (Rushmer et al., 1965; Mullins, 1961). The transducers are fixed to structures such as the walls of a blood vessel or the heart, and the record of changes in transit time is used to measure the fluctuation in distance between these walls.

In our method for measuring bone mass, the sonodistometer is modified to measure velocity of sound in any given medium. To do this the distance between the transducers is kept constant. Therefore, the transit time is reciprocally proportional to the velocity of sound in the medium separating them. The elements of this instrument are shown in figure 1. Two piezeolectric ceramic transducers are fixed rigidly to opposing ends of a U-shaped frame. One transmits a burst of sound which passes through the medium between them and is received by the other. In order to prevent the very substantial absorption of sound by air, the transducers and any substance to be examined are immersed in a tank of water. The transducers are of barium titinate and are shaped so that their natural thickness mode of vibration is 3 megacycles. This high frequency is used to achieve a narrow, well-defined beam so that

good resolution of structures within the tissues examined is obtained.

To produce a sonic vibration in the water, a 900-volt pulse is fed through a pulse transformer to give it the rise time characteristic of the 3-megacycle sinusoidal frequency. This is applied to the transducer to generate an abrupt train of sinusoidal oscillations. The train is dampened so that the total duration of the sonic burst is about 15 μsec. The transmitter is so pulsed 60 times a second; but as the duration of each burst of sound is short, the amount of energy which could be transmitted to a living tissue is small—less than 9 cal per minute even if entirely absorbed.

The converter section, which has been modified from that of Mullins (1961), is shown by figure 2. A voltage ramp is started at the time the transmitter transducer is pulsed and cut off by the signal from the receiver transducer. The properties of this voltage ramp are such that the voltage rises at a constant rate during the time between these two signals. Therefore, the maximum voltage attained is directly proportional to the time between transmission and reception of the pulse. The voltage is stored on a capacitor until the next time the transmitter fires; then the storage capacitor is discharged and a new charge is stored on it by the ramp. The discharged voltage is amplified and fed to a recording system which produces a line that varies position with the intensity of the stored voltage. The results are displayed on an oscilloscope and by a rectilinear recorder.

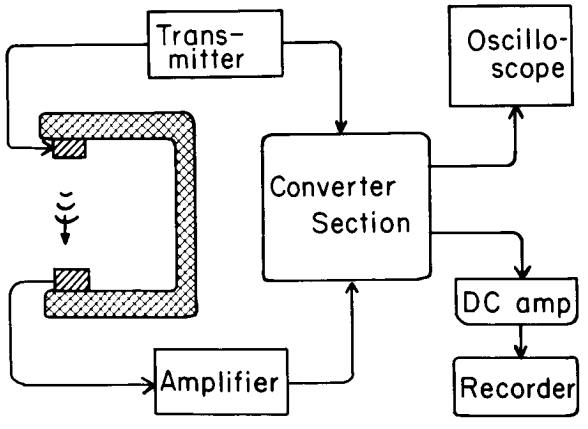

FIGURE 1.—Block diagram of the sonic velocimeter.

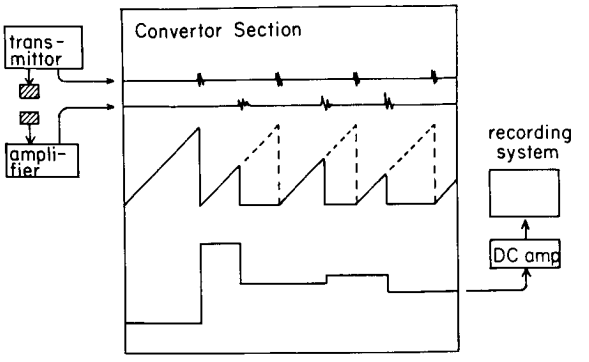

FIGURE 2.—The conversion system used in the sonodistometer and adapted for sonic velocimetry.

There were several problems which had to be solved before the sonodistometer could be used for the measurements described herein. First, to handle a large common mode signal of electrostatic origin from the transmitter transducer, a balanced input was employed. This, together with techniques of containing the electric field in the transmitter crystal, kept the electrostatic coupling effect within usable limits. Second, the output from the receiver crystal varied because of large losses from reflection when the sound beam was passed through irregularly structured material such as trabecular bone. The transmitted signal was 900 volts. When only water was between the transducers, the output from the receiver crystal was usually about 10 volts; but when trabecular bone was interposed, it fell to 100 μV, and sometimes less. Accordingly, it was necessary to use an automatic gain control circuit which variably amplified the signal from the receiver transducer, to achieve approximately the same amplified signal strength, irrespective of the voltage input. While no amplification was necessary for a strong signal, a weak signal might be amplified as much as 10,000 times. The limiting factors are inherent in the amplifiers themselves, and it is a fact that with higher amplification the signal becomes indistinguishable from noise.

This system, which we call the sonic velocimeter, allows measurement of changes in transit time when the signal is passed through a given medium. If the distance between the transducers has been measured, the volocity of

sound in any homogenous tissue can be calcuted. In practice, the distance between the transducers is calculated from a measurement of the transit time of a sonic pulse passed through a reference medium in which the velocity of sound is known.

Slices of bovine cortical or trabecular bone were cut and polished to form rectangles. The dimensions of these segments of bone were measured by a micrometer, the bones weighed and finally (after sonic measurements were finished) ashed in nitric acid and the calcium content determined by titration with versine (Jones and McGuskin, 1964). Other tissues were measured after being packed lightly into a 1-cm lucite cell which contained the transducers in its walls.

To measure changes of transmission time in nonhomogenous structured tissue, the sensing device was passed back and forth at a constant rate over the tissue in a regular geometric pattern, and a continuous record of signal transmission time was made by appropriate recording apparatus. This technique is analogous to that used in radioisotope scanning, where a radiation detector is passed back and forth across the region over-lying a radioisotope accumulating organ, and a graphic record is made of the radioactivity under the detector. The scanning sonic velocimeter is shown in schematic form in figure 3. The frame which holds the transducers is fixed to the detector of a commercially obtained clinical radioisotope scanning device, as shown at the upper left. The crystal transducers, the sample, and a standard are placed in a tank of water in such a way that they are scanned as the frame is moved back and forth, as shown at the upper right. An idealized recording from this path onto a strip chart is shown at the bottom. This indicates a "base line" transmission time when the signal passes through water alone (a), and a change when it passes through the standard bone wedge (b). As the sonic path traverses the sample, there is a change in transmission time indicating soft tissue (c), and an additional change when it passes through soft tissue and bone (d). The difference between transmission time through bone and soft tissues and that through soft tissues alone can be compared to the transmission through a bone chip of known composition. This allows estimation of the mass of calcium in the bone examined. When the dimensions of the bone are known (from an X-ray), its density can be estimated.

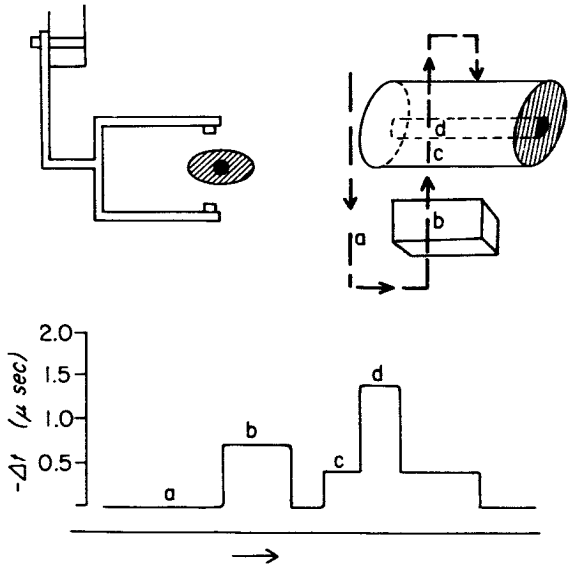

FIGURE 3.—Diagram of the operation of the scanning sonic velocimeter.

CALCULATIONS

It is assumed that the distance between the transducers is fixed, that the velocity of sound in any given medium is constant, that the voltage output is directly proportional to the transit time, and that, when a scan is made, both the transducer frame and chart recorder move at constant rates. Then, when a single medium separates the transducers, $d=t/c$, where d=cm, $t=\mu$sec, and c=cm per μsec. When the velocity is known (reference medium in this work is water at 20°C; $c=0.143$), this can be used to determine d. Thereafter, with d known, c of other media can be determined.

When several media are placed between the transducers, $t=d_1c_1+d_2c_2 \ldots +d_nc_n$, where d_ic_i represent the distance of the sonic path and velocity in the i^{th} medium and $d_1+d_2 \ldots +d_n=d$. This relationship can be used to calculate c_i or d_i if the other factors are known.

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If, in the passage of the sonic pulse through a limb immersed in water, t_1 is time through water, t_2 through skin, t_3 through muscle, t_4 through other soft tissues, and t_5 through bone, the total time through a part of the limb immediately adjacent to the bone $= t' = t_1 + t_2 + t_3 + t_4$, and that through bone $= t' = t_5$. If the distances d_1, d_2, d_3, and d_4 have not changed significantly between the two measurements, the difference in total time measured is t_5. If the velocity of sound in bone is c_5 and has been independently determined, d_5 can be calculated. In theory, this gives the thickness of bone in the sonic path, independent of its shape or state of dispersion (cortical or trabecular). This value can be converted to the mass of bone or calcium in a column of unit area in the path of the sonic beam.

The sonic scan theoretically gives the same information for any linear trace over the limb. The height of the sonic scan is proportional to t and the breadth proportional to the distance the transducers have moved. These together describe an area on the paper. When A_s and A_u are the areas so produced for a standard and an unknown, and M_s and M_u are the amounts of calcium per square centimeter normal to the sonic path, $M_u = A_u\ M_s/A_s$.

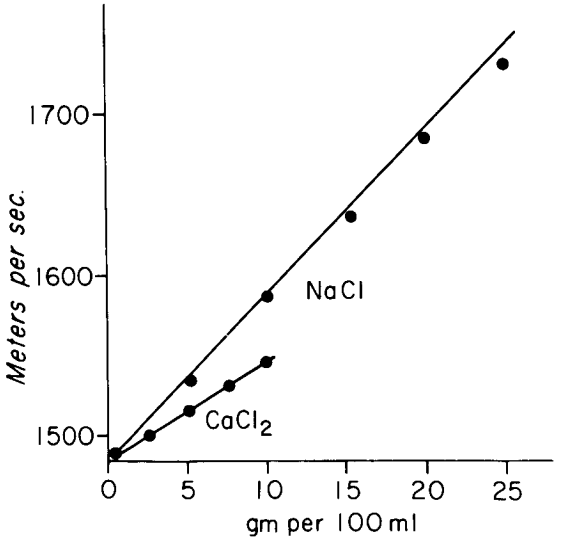

FIGURE 4.—Velocity of sound in solutions of different concentrations of NaCl and $CaCl_2$.

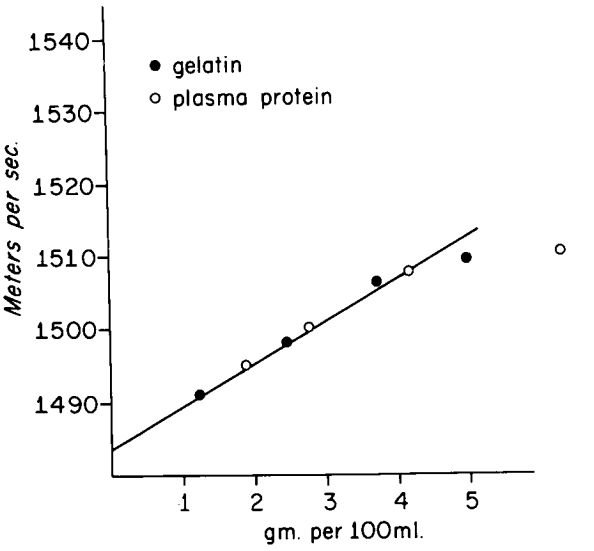

FIGURE 5.—Velocity of sound in different concentrations of gelatin and plasma proteins.

RESULTS

We found the velocity of sound in beef bone, muscle, and fat to be 2880, 1630, and 1540 M per second, respectively. The measurements were made at 20° C and are based upon the assumed velocity of sound in pure water of 1430 M per second (Carlin, 1960). There is variation when such nonhomogenous substances as muscle, fat, and cancellous bone are measured, as would be expected because of the lack of uniformity of the tissues examined. The velocity in fatty tissue is greater than that found in pure fat (1460 M per sec), but this is not surprising, as the tissue contains cell protoplasm as well as fat. It is presumably because of the fat that velocity of sound in fat tissue is less than in muscle.

Figures 4, 5, and 6 show the velocity changes in different concentrations of salt and protein and different mixtures of blood cells and saline. The relationship between protein concentration and velocity was nonlinear at the higher concentrations as could be expected, because velocity is a power function of compressibility and density in liquid media. A linear relationship is found when sound is passed through differing concentrations of erythrocytes; as in this case, the concentration of protein within the cellular

phase is not changed, only the fraction of the medium occupied by cells. The changes in velocity appear to be accounted for by the protein in the red cells.

Figure 7 shows the difference between transit time when the fixed transducers are immersed in water alone and when chips of compact bovine or trabecular bone of different thicknesses are interposed between them. On the abscissa is plotted milligrams of calcium per square centimeter normal to the sonic path, and on the ordinate is the measured decrease in transit time. For cortical bone there was a high correlation between change in transit time and either length of the sonic path through bone or, as plotted in figure 7, mass of calcium in the sonic path. There was a slightly better correlation between transit time and mass, rather than distance, as might be expected because of the variability in calcification of different samples of cortical bone. The coefficient of variation for the entire set of measurements on cortical bone was 5%.

The same accuracy is not seen when chips of trabecular bone are examined. These represent a much more technically difficult measurement because of loss of a large part of the sonic energy, presumably from reflection and formation of shear waves as the beam is passed through the many interphases between bone and marrow. However, even when these technical difficulties are overcome, the resulting measurement fails to predict accurately the calcium content. As indicated in figure 7, the sonic measurement gives a value which usually results in too high a prediction of the mass of calcium present. The possible reasons for this will be commented upon later.

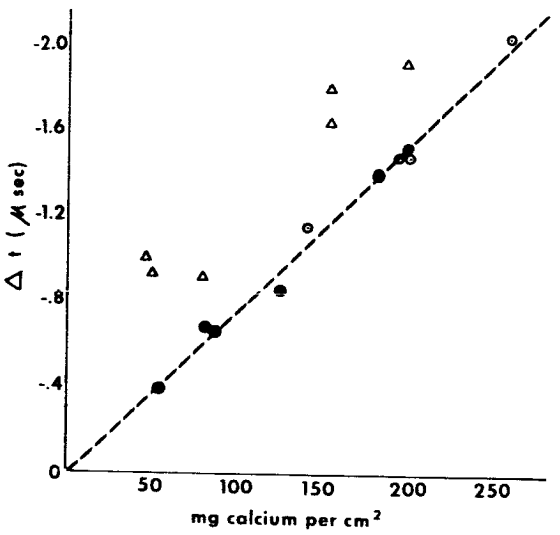

FIGURE 7.—Decrease in transit time from that in water alone when sound is passed through different thicknesses of bone. On the ordinate is the change in transit time (μsec) and on the abscissa the mass of calcium through which the sound passes (mg Ca per cm² normal to the sonic path). Circles give values for cortical bone and triangles those for trabecular bone. Closed circles are measurements made on an early instrument 2 years before those represented by the open figures.

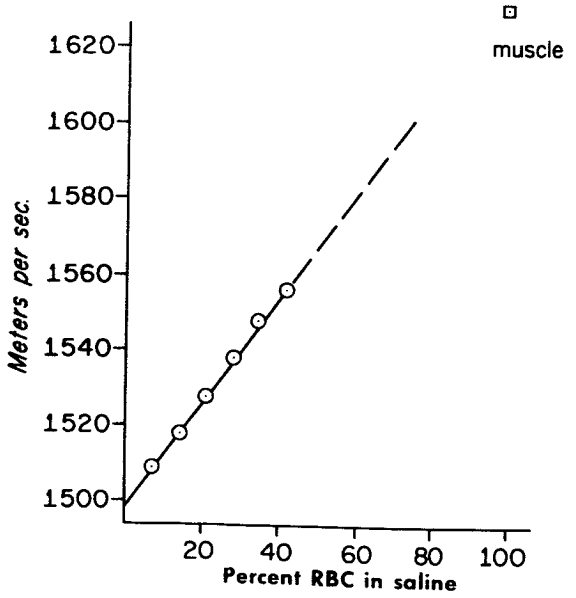

FIGURE 6.—Velocity of sound in different mixtures of human erythrocytes and 0.9% NaCl. Also shown is the velocity of sound in a fully cellular tissue (bovine muscle).

In order to determine the accuracy with which the sonic scanning velocimeter can be used to predict the mass of bone present within an intact limb, a rabbit femur was scanned 0.5 cm from its proximal end and each centimeter below this point for 7 cm. A typical result (at 2.5 cm from the proximal end) is shown in figure 8. The area due to bone could easily be identified

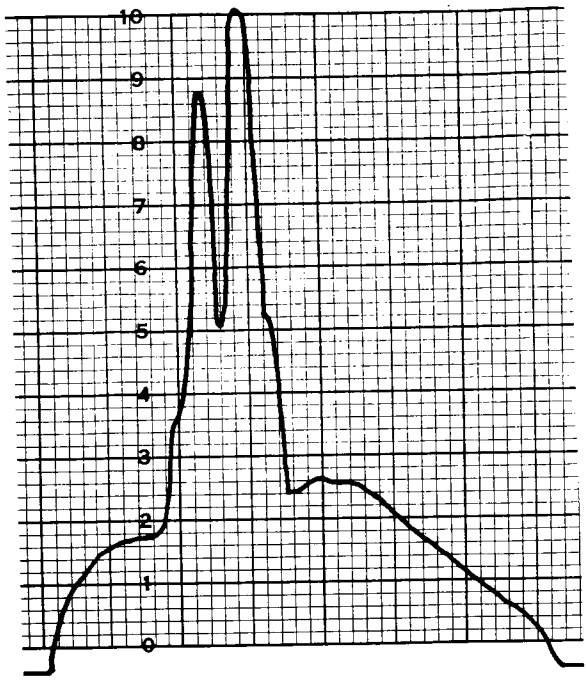

FIGURE 8.—Sonic scan of a rabbit thigh.

in all the scans, but the two most proximal were very irregular because of reflections from the different joint surfaces and could not be used. The soft tissues were then removed and the femur again scanned at the same point. The areas due to bone of all but the two proximal scans were determined by planimetry and are given in table I. A rectangle of bovine cortical bone, which served as a standard, was carried through these same steps and its known calcium content used to calculate the mass of calcium present in the path of the sonic scans. The rabbit femur was then divided into 1-cm segments, ashed, and the amount of calcium measured as also shown in table I. There was no significant difference between the area of scans of the intact and stripped bone, the small changes observed being accounted for by the improbability that the two scans were exactly in the same place. The standard deviation of the difference between predicted and found values was 6.6 mg calcium, and the coefficient of variation, 4.2%.

Figure 9 shows the scan of a stripped bovine femur. It is easy to identify in this and in

TABLE I.—*Ultrasonic and Chemical Measurements on a Rabbit Femur*

Position of scan (cm from head)	Area of bone profile in sonic scan (cm²)		Mg calcium per cm segment of bone		
	Intact limb	Bone only	Predicted from scan	Found	Difference
2.5	26. 8	26. 5	174	187	13
3.5	26. 8	27. 5	181	177	−4
4.5	28. 5	26. 5	174	187	13
5.5	24. 5	24. 0	158	169	11
6.5	23. 0	22. 0	149	143	−2
7.5	----	18. 0	118	125	7
Standard (bovine cortex)	----	39. 5	------	260	----

figure 8 the part of the scan which results from the passage of the transducers over the middle part of the bone where the beam passes perpendicularly through the cortex, as compared to the lateral part of the scan, where it passes tangentially through a greater distance of bone.

DISCUSSION

Several applications of ultrasound to measurement of tissue structure *in situ* have been made. Most work has been based upon reception of a reflection of sound from some interphase within the tissue examined. Some early work was based upon absorption of sound in tissues (Carlin, 1960; Newell, 1963), and a few measurements of transit time through tissue have been made (Ludwig, 1950); but these latter methods proved technically difficult and were abandoned. We have taken advantage of recent technological developments which made the transmission measurement more feasible.

This system shows promise of accurate measurement of the mass of the tubular portions of the bones of the limbs, and can probably be used also to determine some properties of the soft tissues. Although this may result in some worthwhile applications, it would be very desirable if measurements of the mass of trabec-

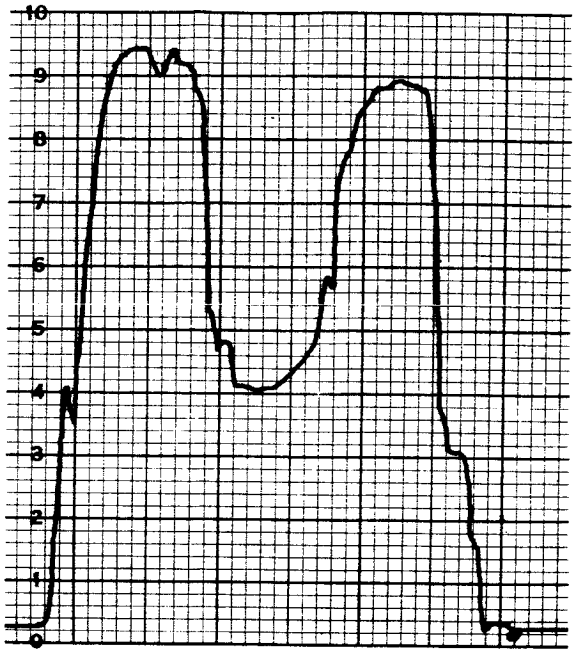

FIGURE 9.—Scan of a beef femur from which the soft tissues had been removed.

ular bone could be carried out as well. Several factors which may significantly influence this measurement are now being evaluated. There is much greater absorption of sound in trabecular bone than in other tissues, presumably largely because of reflections of sound as it passes through the many irregularly arranged interphases. This can possibly be overcome by increasing the signal strength, by reducing the noise (and thereby allowing more amplification of the current from the receiver transducer), or by altering the frequency. We have used a high frequency to obtain a narrow beam, but this can be achieved by other means, and sound of a lower frequency is not so greatly absorbed (Carlin, 1960).

The other difficulty is that, in an irregularly dispersed system such as trabecular bone, some portion of the sound may pass through a particularly dense portion of bone and thus arrive sooner at the receiver transducer than it would have otherwise. This would cut off the voltage ramp early and the velocimeter would indicate a greater amount of bone than was present on the average in the path of the sonic beam. When the cross section of the beam is relatively large compared to the condensations of bone over which the transducers pass (as is the case for trabecular bone), this effect could cause the sonic scan to indicate more bone than was present. Attempts are now in progress to reduce this difficulty through use of transducers specially shaped to give an intense beam of only a few millimeters in diameter.

A theoretically more difficult problem is illustrated by figure 10. The sound beam need not go in a straight line through bone, as shown to the left. Instead, it may take a circuitous course in a trabecular system, as shown on the right, so that it passes through solid bone for a greater portion of its transit than it would if it went in a straight line. This would result in the sonic scan giving too high an estimate of the mass of trabecular bone. Further work will be required to determine whether one or both of these factors, or others not yet identified, are the cause of the inaccurate measurements of trabecular bone mass.

The potential advantages of this system are its precision, safety, speed of operation, and, in respect to cortical bone, its accuracy. At present, we can scan the human forearm, wrist, and fingers and presume that the same could be done for some bones of the leg. The inability to accurately estimate the amount of calcium in trabecular bone and the fact that absorption of power may be too great if the beam is passed through a thick part of the body make it seem unlikely that the present instrument can be applied to measurement of vertebral mass.

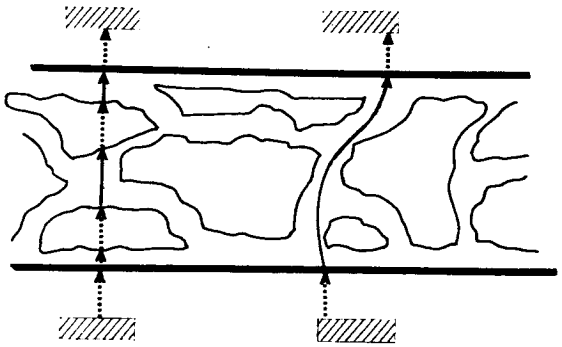

FIGURE 10.—Comparison of possible paths of the sonic pulse through trabecular bone.

REFERENCES

CARLIN, B.: Ultrasonics. Second ed. McGraw-Hill Book Co., New York, 1960.

JONES, J. D.; and MCGUSKIN, W. F.: Complexometric Titration of Calcium and Magnesium by a Semiautomated Procedure. Clin. Chem. vol. 10, 1964, p. 767.

LUDWIG, G. D.: The Velocity of Sound through Tissues and the Acoustic Impedance in Tissues. J. Acoust. Soc. Am., vol. 22, 1950, p. 862.

MULLINS, G. L.: Sonodistometer. Digest of the 4th International Conference on Medical Electronics, New York, 1961, p. 70.

NEWELL, J. A.: Ultrasonics in medicine. Phys. Med. Biol., vol. 8, 1963, p. 241.

RICH, C.; KLINK, E. J.; MULLINS, G. L.; and GRAHAM, C. B.: Sonic Measurement of Bone Mass. J. Clin. Invest., vol. 42, 1963, p. 970.

RUSHMER, R. F.; FRANKLIN, D. E.; and ELLIS, R. E.: Left Ventricular Dimensions Recorded by Sonocardiometry. Circ. Res., vol. 4, 1956, p. 684.

COMMENTS

Dr. MALETSKOS. What size area of bone did you use?

Dr. RICH. Two or 3 cm x 1 or 2 cm were used.

Dr. MALETSKOS. I was trying to get the relationship between the area of crystal and the area of the bone.

Dr. RICH. The crystal is only 0.5 cm in diameter or less. This can be varied. As I will explain later, we would like to have a point source, but we must get some power out of the thing. This is a problem that we are embroiled with right now.

Dr. URIST. Was that calcium hydroxyapatite, bone ash, or bone?

Dr. RICH. It was bone. These are carefully milled rectangles of cortical bone. These are the same data replotted (fig. 7) and these obscure open circles are a set of new determinations done 2½ years later with an entirely different machine and different chips of bone.

What this shows is that the velocity of sound has not changed in the last 2 years in bovine bone, not too striking a discovery. This entire set of data simply indicates something about our present precision.

If you divide one of these by the other you come up with a figure of 0.131 ± 0.0066, which gives a coefficient of variation of 5%. This is not by any means the result of much effort at precision. The factors in instrumentation that we are now using could be controlled further.

Dr. LANZL. Could you elaborate on what A is?

Dr. RICH. A is the area of sonic profile in square centimeters of the bone, in this case the average of all segments.

Dr. LANZL. Is it the profile at the bone?

Dr. RICH. As the machine is passed over the bone, we get an area in which the height is proportional, we trust, to the amount of calcium present. The distance is a function of the length of the dimension that we scan, so that the area should be proportional to the amount of calcium per unit area in the path of the bone.

What I was trying to indicate was that the values predict fairly well from one part to another of this bone, one segment of which was trabecular. I am not quite sure why it was not off, but it was not. We can use, in this case and in others that we have done, the scan of a standard bone chip to predict the amount of calcium that we would have found. In other words, this is what one wishes to do in bone mass measurement.

Dr. LANZL. Could you say anything about the energy cross section of your beam at the bone interface?

Dr. RICH. No, I really could not. We start out with 900 volts and end up in the beam with only 2 to 10 volts, depending on the amount of soft tissue present. It is quite variable. We are really not dependent on the energy here so much as the velocity.

Dr. LANZL. I was trying to get at what was the profile of the beam at the bone, the dispersion.

Dr. RICH. We have measured this only in water. I am sure it is altered as it goes through a complex tissue. The measurement of it is rather complicated; and from our present point of view, since we are trying to do away with this unevenness in this profile, not of first order of interest to us.

Dr. URIST. Would you say the more homogenous the solid phase, the more reproducible?

Dr. RICH. Yes. We do not have any trouble with the homogeneous solid phase. The problem is the interface in the trabecular bone.

Dr. URIST. I assume you have run it down the long axis.

Dr. RICH. Yes, we turn our cortical bone at 90°. Within the precision of the machine we cannot see the difference. There is reason, since the elastic properties of the medium also affect the velocity, that you

might see the difference, but I think you need greater precision than we have. For example, we just measure the bone within 0.1 mm. One centimeter bone is a fairly gross measurement for the type of thing we are getting at. Our present view is that this still has a number of quite severe technical problems. Although I realize that the data I showed had coefficients of variation around 4 or 5%, these are due to problems that one can't control.

I might say the speed of the scan, certainly the safety, and we believe the accuracy in cortical bone, even when fairly deeply buried in soft tissues, is of a rather encouraging order.

I think it is too early to comment as to whether trabecular bone will yield to our efforts. Our future plans are to continue to attack some of these technical details, and also to apply this to a study of human bones in the limb.

Dr. CAMERON. Have you used it on limbs yet?

Dr. RICH. Yes, we use it on our own limbs. This is an interesting thing, because we have great trouble with the little cube of bovine trabecular bone when we take it out and isolate it, but we can get through the wrist. We can scan right down the hand, yet I don't understand it. It may be that the sound is passing along the cortical envelope and we are not getting an accurate measurement. I don't know yet. It is a preliminary report.

Dr. NORDIN. Are you operating at a different wavelength from the one ordinarily used for reflection purposes?

Dr. RICH. Yes, but this is 3 megacycles.

Dr. NORDIN. What is the normal figure?

Dr. CAMERON. I think it is about 1 megacycle.

Dr. NORDIN. These are used for sound for reflection?

Dr. RICH. For reflection, yes.

Dr. NORDIN. Could you tell us what advantage you thought this would have over other forms of radiation when you started and what your conclusion is? I mean relative to X-radiation and γ-radiation. How does this compare?

Dr. RICH. Before we realized that we were really in hot water with trabecular bone, this method seemed to me worthwhile in that it offered a high degree of precision, far higher than what the radiographic techniques in general were giving at that time. Now the latter appear to have been improved, but all may have a limiting precision of about the same order.

We thought, since the principles of measurement are entirely different, there could be different applications, different problems, perhaps different areas of optimal use. To put it a little differently, usually it is worthwhile exploring new approaches.

Dr. NORDIN. Does not this trabecular bone perhaps come down to a matter of frequency? You are getting some reflection, aren't you?

Dr. RICH. Reflection is not too much of a problem. The wavelength of 3 megacycles is probably in the same general range of the dimension of a trabecular limb. There is theoretical reason for believing that the lower frequency probably will present other problems, while lessening problems with the graininess, the reflection in shear wave formation in trabecular bone. We might get other sources of loss of energy. There is confusion when the frequency is too long.

This is definitely one of the things that we are investigating. The speed of our investigation has been somewhat delayed because of instrumentation.

Dr. COHEN. I am curious about the effect of geometry on this transmission. Suppose I had a cylinder of cortical bone, and I measured it one way along the axis and then transversely. Let us further say the total dimension stayed within the beam. Would you expect a difference in time of the leading edge arriving at your detector?

Dr. RICH. Not theoretically. My experience with this system has led me not to predict very much.

Dr. COHEN. If you have a fast velocity path, you would get between points sooner.

Dr. RICH. There is no question about it.

Dr. COHEN. If it is a question of the amount of bone measured against a cube, there would be some uncertainty.

Dr. RICH. I do not think there is any, with the exception of trabecular bone. I think we may be getting a facilitated path through condensation of trabeculae. We always measure the most bone, not the average bone, between the transducers.

Dr. LANZL. Have you taken your sample and moved it closer to the transmitter or made some sort of plot of moving the sample in between your transmitter and receiver?

Dr. RICH. Yes, we have. There are a number of problems which I did not touch on too much. The transmission signal strength is another thing that we can and do modulate. This has considerable influence. Do you recall that diagram I had indicating the irregular shape? If the signal strength is low, these things do not get transmitted through a large amount of tissue, whereas if the signal strength is greatly increased, you start seeing it. There are other examples of the influence of signal strength. With regard to your question more specifically, we pile samples on top of each other; this makes no difference. We can move the samples away from or toward the transducers, the transreceiver, or transmitter-transducer without difference except that if we move it close in, then this has the effect of making these things more troublesome.

Dr. LANZL. In your understanding of the trabecular bone, you are taking the bone and stretching it out in a sense. Maybe you are not as bad off as you might think?

Dr. RICH. I do not know if that formulation I made on the slide with the circuitous route is correct. Theoretically, if it were simply a homogeneous dispersion, little round globs of bone in soft tissue, then I do not think we would be in any trouble so long as we get

the signal through. We have tested this in various ways by making solutions of gelatin, etc. to test the fact that a given quantity of material dispersed gives us much the same result as the same amount of material compacted.

Dr. LANZL. How about lamellae if you again take a sheet?

Dr. RICH. That is the point. That is what is happening in trabecular bone. This is an analog of a trabecular bone. If we take a cortical bone and drill a hole in it, if that hole is small, we do not see it. We have not done what you have just suggested. I have no doubt whatsoever that if this is the crystal size and if we put a sample of lamellar bone in like this or something looking like a toothbrush or comb, it will look to the crystals as if it were solid. If you turn at right angles, you get the result you should. The number of interfaces at right angles is no problem at all. If it is a straight line path, and when the transmission signal is intense enough, then the first signal that is transmitted down trips the mechanism. This is something we cannot get away from, other than by reducing the size or the cross sectional area of the signal which we are trying to do now by modulating the transmission strength.

Dr. ROCKOFF. You could theoretically oscillate the bone at a very, very high frequency. Would you improve then at all the certainty that the sonics are going through more diffusely?

Dr. RICH. I really do not know. I doubt if this would be practical. There is little reason to believe that the whole bone oscillates at this frequency. This is quite a high frequency, 3 megacycles. We have thought about rotating the bone rather than our transreceivers. What I thought you were going to ask about is modulating the frequency. This is a real technical problem because this is a pulse technique. Once you get into an FM technique, it is very difficult.

Dr. MALETSKOS. The kind of oscillation Dr. Rockoff is talking about would be of such frequency that mechanically you could not get this?

Dr. RICH. I think we are more interested in trying to get a very well defined sharp beam. Lower frequencies may be helpful.

APPLICATION OF TECHNIQUES IN HUMAN STUDIES

HERBERT POLLACK, *Chairman*

Rates of Involution of Vertebrae and Femur in Aging

JAMES S. ARNOLD AND MURRAY H. BARTLEY
Sumerlin Memorial Pathology Laboratory

During the last few days we have heard Dr. Nordin and Dr. Mack discuss the X-ray quantitation of vertebral mineralization in living patients. At this time, I would like to demonstrate what the actual ash per unit volume in vertebral centrums is, as measured in autopsy specimens.

For some years now we have been cutting rectangular cubes from the mid-central area of the lumbar vertebra, measuring the external dimensions with a caliper, and determining the mineral per unit volume by ashing. In figure 1 the ash per cubic centimeter of vetebral medullary tissue is plotted as a function of age. The round dots represent the females and the squares, males. It is apparent that during the first 20 years of life there is a progressive increase in the ash concentration for both sexes, and a progressive decrease in both sexes after the age of 30. The levels of ash per cubic centimeter in females are always somewhat lower than those of the males, but the shape and the height of the curves are comparable. The data in figure 1 are derived entirely from cases dying traumatic deaths, or from ruptured cerebral or abdominal aneurysms. Myocardial infarcts were not included, as there is usually a question of chronic heart disease associated with these cases.

Figure 2 contains similar data of ash per cubic centimeter vertebral medullary tissue as a function of age for a variety of chronic disease states. Here we see a similar progressive decrease in vertebral ash concentration as a function of age. In addition, there are a definite

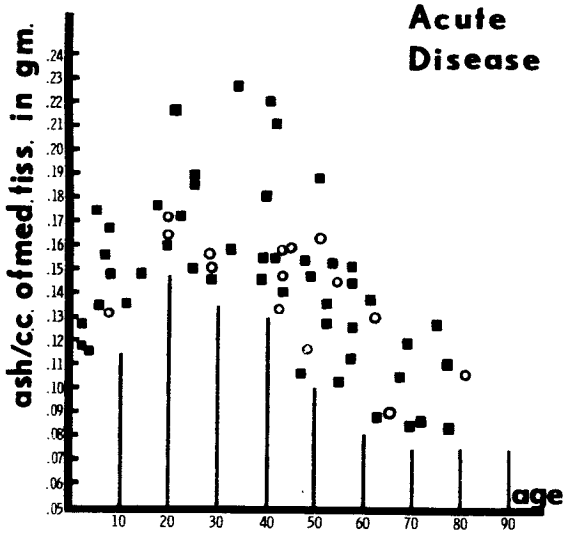

FIGURE 1.—Ash/c.c. of vertebral medullary tissue vs. age.

number of cases with an increased ash per cubic centimeter of vertebral tissue. These cases correspond principally to cases of chronic renal disease and are therefore thought to represent osteosclerosis, which is not readily detected by the usual techniques of pathologic examination. The female cases, particularly, show considerable reduction in ash per cubic centimeter in the older ages, below those seen in the acute cases. Thus, there is indication that there is a definite reduction in vertebral mineralization in chronic, debilitating disease, and more conspicuously so in females.

Dr. Nordin has earlier told us of his im-

pression that some cases of vertebral osteoporosis are associated with narrowing of the femoral cortical bone, while others are not. In autopsy material comparable data are seen. In figure 3 the thickness of femoral diaphyseal cortical bone is plotted against the ash per cubic centimeter of vertebral medullary tissue. Again the females are plotted as circles and the males as squares. The cases in the lower left-hand corner of the graph represent cases where both the femoral and vertebral atrophy have taken place, while the cases on the upper left-hand side correspond to cases where vertebral atrophy occurred without apparent atrophy of the femoral cortex. Virtually no cases are present in the lower right side of the graph, indicating that there are virtually no cases where the femoral cortex atrophies in the presence of a normal degree of vertebral mineralization.

The female cases generally show considerable reduction in the femoral cortical thickness for comparable amounts of vertebral mineralization. This seems to be a consistent finding—that the females show considerable cortical thinning as compared with men, throughout the age span, and more strikingly in old age. When cortical thickness is plotted against age, the females show progressive atrophy, or thinning,

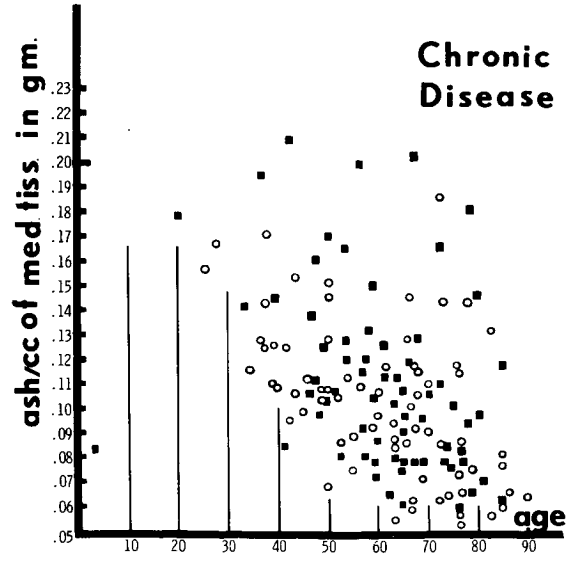

FIGURE 2.—Ash/c.c. of vertebral medullary tissue vs. age.

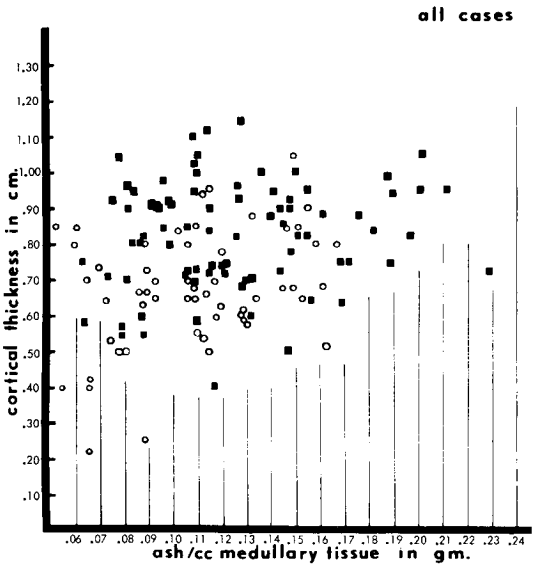

FIGURE 3.—Maximum femoral cortical thickness vs. ash/c.c. vertebral medullary tissue.

of the femoral cortex as a function of age, while the males seem to be divided into two populations. The largest population shows no change in femoral cortical thickness as a function of age, while a smaller population appears to atrophy in a manner comparable to that of the female population. It appears that there is vertebral atrophy in virtually everybody, starting at about 30 years of age, reaching a baseline someplace in the vicinity of 90+ years of age. This is true for both males and females. The females appear, throughout life, to have slightly lower values than males. The femoral cortical bone is progressively thinning throughout life in the females, while it appears to be constant in thickness in the majority of males. It would appear that the reason that the female aging population gets into trouble, from the standpoint of vertebral and femoral fractures, is that the vertebral atrophy occurs more rapidly. Also the femoral atrophy begins with a thinner starting value and occurs more rapidly in females. From the standpoint of vertebral mineralization, the difference between men and women then appears to be due chiefly to the difference in behavior of cortical bone. Female bones atrophy, while male bones do not.

COMMENTS

Dr. NORDIN. How did you measure the volume of the vertebral sections?

Dr. ARNOLD. It was done with a caliper. I cut them into cubes and measured the cubes. Initially I fixed the bone cubes in formaldehyde and calculated by Archimedes' principle, but the other technique is much easier.

Dr. NORDIN. Have you made any attempt to correct for the quite substantial variation which exists from one part of the vertebral body to the other? You notice that the central cubic centimeter is somewhat more porous and more vascular than the peripheral part.

Dr. ARNOLD. Yes, they are all taken pericentrally. I cut the vertebra right down the middle, and then I measure a piece on each side.

Dr. NORDIN. That is why your values are a little on the low side as compared to those of other researchers. Yours are right for the central cube of the vertebral body, but they would not be right for the whole vertebral body.

Dr. ARNOLD. That is right. These are without the cortical bone. We have now more than 300 cases.

Dr. RICH. Do I notice that there is a sharp break in the drop in the density in both males and females after menopause?

Dr. ARNOLD. Or the age corresponding to this. This is taking all varieties of chronic disease that come to the autopsy table. The high ones are what you would call osteosclerosis; all of these individuals have chronic renal disease.

Dr. SMITH. Did you say which vertebral body you were taking, in view of your comment yesterday about which ones seem to be involved?

Dr. ARNOLD. I have been measuring usually the first or third lumbar vertebra. Some of these data have been recorded over 5 years. I have done different experiments and moved around, but within the first to the third lumbar vertebra. There is no relative difference from the 11th thoracic vertebra down through the fourth lumbar as far as I can determine. In many of these cases I have measured three different vertebrae and the values are virtually interchangeable.

Now, these values are lower in the chronic disease case; in other words, there are more osteoporotics. I would call anything osteoporotic which fell below point zero eight or certainly point zero seven. But here again the cut-off value is very difficult because you may have compression fractures all the way up to point zero nine, but generally you do not see them until below point zero six. In other words, the point at which patients have compression fractures does not seem to be exclusively a matter of the mineral concentration in the vertebrae.

Dr. SMITH. In general, compression fractures relate to osteoporosis but not necessarily.

Dr. ARNOLD. That is right.

Dr. NORDIN. Another point is that some of the strength of the vertebral body, no one knows quite how much, is undoubtedly due to the cortex. And you are measuring the central cube of trabecular bone?

Dr. ARNOLD. That is right, exactly.

Dr. URIST. This is physiological reduction of bone mass. It is proportional to the reduction in muscle mass. Bone mass and muscle mass are reduced together, but you get just as much bone reduction as you do muscle atrophy. This is something that is in a sense time dependent, for which we use the word aging. We have no idea what this word means, but in a vague way this is a rate of reduction of bone mass with time. It is normal in the United States and, as Dr. Trotter showed, there is a curve for this for Negro males, another curve for Negro females, and another for white males.

This is a specific thing. Where the physiologic picture breaks off and becomes the disease entity, this is what seems vague to me, and I am not clear on this yet, but I think that one is a disease and one is a physiologic process.

Dr. WHEDON. The prevalence of diabetes increases considerably with age, particularly after the age of 40, but we do not talk about physiological diabetes with aging. Would you?

Dr. URIST. I would not have any objection to it if you did.

Dr. WHEDON. My comment is by way of objecting to the whole concept of calling an abnormality physiological.

Dr. NORDIN. I think that one is always up against this problem: When does an abnormality become a disease? I think we should face this in a discussion of osteoporosis, about which we talk glibly. Everybody at this meeting has used the term without facing up to the fact that it is very difficult to define what we mean by it. I would suggest that if one were to look at it in the sense that you can define an abnormality in some sort of statistical terms, and you can define disease in some sort of clinical terms, then you might be able to break the situation down into people who have normal bones, and people who have the clinical consequences attached to it.

For most parameters, of course, we have the marvelous Gaussian distribution of two standard deviations about the mean. It appears that there is a clinical disorder of reduced bone volume which is very common without any corresponding clinical disorder of increased bone volume. So, it is not possible to take the mean amount of bone and two standard deviations on either side and say that is the normal range. It has a grossly skew distribution. We tried to tackle this problem by collecting iliac samples from the postmortem room and grading them according to the amount of bone present per unit volume. We found that the frequency distribution histogram had a typical peak representing the normal population and an abnormal tail on one side. Separation of the normal

from the abnormal takes place at a figure of approximately 16% of the iliac crest sample occupied by bone. That figure has been reached quite independently by the Dutch, who published recently a series of observations on really normal people, volunteers, students, etc., and reported that the low limit of normal in the iliac crest was 17%. There is also a series of data published which comes out to exactly the same figure, a weight to volume ratio of 1 in 6 which is almost exactly the same as this. We have found in 275 consecutive autopsies that 23% of the women and 12% of the men were below that figure.

Dr. RICH. What were their ages?

Dr. NORDIN. They were predominantly elderly. You cannot call them a cross section of the aging population because people who are dying in hospitals are not considered a good cross section. The figure cannot be far off, inasmuch as Caldwell and Collins, in Sheffield did the same thing on post mortems using specific gravity of the bone specimens. They used specific gravity of little cubes cut out of the vertebral body as Arnold did. They decided osteoporosis was present at a specific gravity of bone of 1.050 in a distribution of this kind. On that basis they obtained almost exactly the same figures as we did.

Now as to when this becomes a *disease*, it seems to me that this is very much easier to decide. You can take biconcavity or compression, or you can take incidence of fractures or any number of parameters. The difficulty I think is not in defining the disease but in defining the point at which normality becomes abnormality.

Dr. URIST. At this end of that tail you never see one fracture but a number of fractures, and if you make an X-ray examination of the upper dorsal spine you will see wedging of vertebral bodies; those are microscopic compression fractures. They are always associated with some ballooning of the disk, calcified aorta, and a thin cortex, and there is a spectrum.

In my definition I require all four or five of these indices before I call it a disease. Women are the ones that are most likely to get this disease; they have the longest longevity and the best health. Usually they do not have other diseases; the longest lived mammal is a white woman.

Dr. MALETSKOS. It is clear that physicians have the same problem that a physicist has. They do not know when the signal has come out of the background.

Dr. GARN. If these things weren't happening with age, we would not be dying. I do not think we have to worry really about whether we must use age as a reference standard, but we should merely observe that below certain amounts of bone, regardless of age, the probability of fractures increases. I think that we are all interested, regardless of our age, in not breaking our bones when we step out of the door. I believe that that is what we are trying to find out about here.

Dr. SCHRAER. I would like to go back to the problem of densitometry by X-ray, monochromatic, and perhaps the ultrasonic methods. We have heard all these methods discussed. As I see it now, the film method, as it now stands, is as good as anything that has been discussed up to this point. However, the monochromatic method seems more promising.

Dr. RICH. I hate to get into this fray over aging and osteoporosis, but it seems to me that we are fixing on a common very gross parameter, that is, loss of bone density to identify a process which undoubtedly goes on with age and which probably is a composite of several other pathological processes that give the same picture. I think that the confusion, the difference of opinion that many people hold probably may stem from the fact that we have not developed the ability to look at and understand osteoporosis, and we do not have good diagnostic criteria.

I think, Dr. Urist, that you are way off when you won't accept a person with osteoporosis until she has so much vertebral collapse that her skin is draped around her hips.

Dr. URIST. I said that I recognize osteoporosis as a disease if there is a combination of signs.

Dr. RICH. Can't you believe that some people with this disease do not have these grotesque manifestations of it, just as, for instance, you can have an aortic aneurysm without a rupture?

Dr. URIST. You can have an aortic aneurysm without a rupture, but I do not think you see this disease that we call osteoporosis unless you have a spectrum of changes throughout the whole spinal column, throughout the whole anatomy of the patient's skeletal system. You cannot select bone density as one criterion, because more than bone density is involved. The bones are not only reduced in density but they are reduced in quality; they are reduced in form. When you see the change in a bone, this does not mean that there has been a quantitative reduction in bone mass as though you cut out a piece of bone. You know very well that the whole structure of the bone changes, horizontal trabeculae absorbed, and the vertical ones remain. When you get biconcavity, here is another good example. It does not mean that the cortical end plates have fractures because the bone is insufficient. It means that throughout a period of years the bone structure has been remodeled to make this shape. In other words, a dynamic cellular process has produced these changes.

Dr. RICH. If your hesitancy in making a diagnosis of osteoporosis is simply because you feel the diagnostic criteria are very poor and the clinical ones are lacking, then we are in agreement.

Dr. URIST. I think they are poor.

Dr. WHEDON. On the other hand, when you say you do not have osteoporosis until the end result, I think we are wasting time at the end of a dead-end street; we are going nowhere, and we will never get to the point where we can diagnose osteoporosis early enough to treat or prevent further bone loss. In your criteria your patient does not have diabetes until he has had

acidosis a dozen times. The trend in diabetic diagnosis, however, is to find ways to detect pathologic physiology in the early state called "prediabetes." We have to look for osteoporosis well in advance of multiple fractures. When the patient has demineralization of the skeleton that is obvious to the radiologist with densitometry techniques, he has osteoporosis. We do not have to wait for him to fracture and go into a cast before we say he has osteoporosis. Early diagnosis is one of the purposes of developing a sound densitometric technique.

Dr. MALETSKOS. I would like to make a few comments on the techniques discussed yesterday and today. I want to mention two other methods. One is a device called an isodensitometer which is an automatic device for plotting density in such a way that you plot density increments. This can result in a plot that looks like a contour map, and the scale at which this is done can be controlled. You can take an area of bone on an X-ray film and expand it as much as you want on paper, and at the same time draw in the contour lines of isodensity. This may have immediate application, and perhaps none of you know such a device exists, but it may be useful to you.

Dr. URIST. Equal lines of equal density are plotted. You can visualize the contour by seeing the coding system put on.

Dr. GARN. It is available as an attachment and can be obtained from National Instrument Laboratories, Rockville, Md.

Dr. MALETSKOS. A second technique, on which I have been working, is the use of neutron activation analysis in determining calcium *in vivo*. You are familiar with neutron activation analysis because you can do it on a sample by putting a sample in a reactor, activating it with neutrons, and counting the activity that is formed. By proper arrangement of the experiment, you can do this on a human being without excessively irradiating him. You can do this not only with a reactor beam but even with a neutron generator. I have been working on this phase, trying to do it by measuring the calcium in a part of a bone, say the lining of a finger bone. What you measure is literally calcium. This is a first approach to getting at the chemical composition of a particular material. The great advantage of this technique is that it measures calcium. The calcium concentration of soft tissue is negligible, so you measure calcium in bone.

A group in Britain have carried this one step further and have used a neutron generator to safely irradiate at least two human beings. They are using the technique to study body composition. They can measure the sodium and chlorine and claim they can measure the calcium also. There is no doubt that this is going to be a useful technique in the very near future. It will be used for determination of body composition of a few select elements and, in addition, for trace elements. It can be used also for doing some elegant tracer techniques by making the radioactive material radioactive *in situ* as it normally occurs, rather than by injecting it and waiting for it to redistribute, as it may, in a very short period of time. This whole field I have been calling *in vivo* neutron activation, and it is somthing we might examine in the future.

Dr. WHEDON. I heard you make a presentation last summer at the Gordon Conference on this technique. You cited certain advantages and certain disadvantages. I came away from your presentation feeling rather pessimistic about the future. Did I misunderstand you or has something happened in the intervening 6 or 8 months?

Dr. MALETSKOS. If I gave a pessimistic view that was my intention. The technique is not easy and I was trying to keep from making it sound as if it were a cinch. There are a great many physical problems involved in arriving at the answer which is believable, even with X-ray densitometry and with the use of isotopes. The difficulties that occur in these fields where both medical physicists and medical biologists are involved is that each looks at the problems strictly from his own point of view and their ideas no not meet. I was trying to measure mass of calcium per unit volume of bone. I was carrying it one step further. I wanted to be sure that I had a uniform neutron flux throughout a large volume. At the time I gave the talk I was not sure I could get it. Now I am a good deal more confident.

Experimenters in Britain claim uniform neutron flux through the body. This is one of the points where we differ about what the experiment really does. I think they are going to get it eventually and whoever joins in the work will eventually get it.

Dr. LANZL. What energy neutrons are you using? If you are using slow neutrons, you would not be able to get a very uniform distribution. The relaxation length is about 7 or 8 cm; therefore you could not get a uniform distribution in the body, even though in air you might have a rather uniform flux.

Dr. MALETSKOS. There are various ways of doing this with slow neutrons and with fast neutrons. For calcium it will work very well and will not be influenced by the fact that other constituents of the body will become active, namely, sodium and chlorine, because the energy of the calcium is so high it can be easily delineated.

One other comment. With regard to calibration, some of the speakers have been reluctant to talk either about a mass of bone or composition of bone. In some of the work actual bone pieces have been used as a standard. I think it may be worthwhile to consider what might be a standard bone material. This can be done by combining a series of elements in a plastic matrix, much as I have done in our case with calcium carbonate in lucite. You can make a uniform distribution and you can machine it into any shape. Something that approaches closely the normal chemical

composition of bone would be a material that we should consider to which we could refer all our measurements.

In my particular case, I am interested in activating calcium; therefore I was using calcium carbonate because nothing else would be activated. In other techniques that have been described this could not be done, and other material would have to be added. I would not suggest that you take just bone, clean it with ethylene diamine, powder it, and combine it with lucite, because there might be variations from bone to bone. A standard chemical agent should be used.

In conclusion, you will note that the field of densitometry is growing and expanding at a fast pace. The "eye" was the densitometer and interpreter for a long time. X-ray densitometry has been introduced. This has increased sensitivity and accuracy and is holding its own at the moment. Many other techniques are being examined. We are introducing more variables; therefore we will be able to get more answers. At the same time we are paying a higher price because the field is becoming more and more complicated. As a result, physicians and biologists will have to ask different types of questions because they will be able to get more information and they will be able to dig a little bit deeper. We are in a growing phase not only from the physical point of view but from a biological, medical point of view. The last workshop did not offer much when compared with today's findings.

The Application of Measurements of Bone Volume and Spinal Density

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General Infirmary, University of Leeds

We have applied the measurements of cortical thickness, biconcavity, and spinal density to patients referred with backache and suspected osteoporosis, to a series of cases of steatorrhoea, and to patients on cortico-steroid therapy. Some of the results are reported in this paper.

VISUAL ASSESSMENT AND DENSITOMETRY

To test the clinical significance of relative vertebral density (R.V.D.), we have compared it with visual assessment of films as shown in figure 1. We classified 119 unselected films as

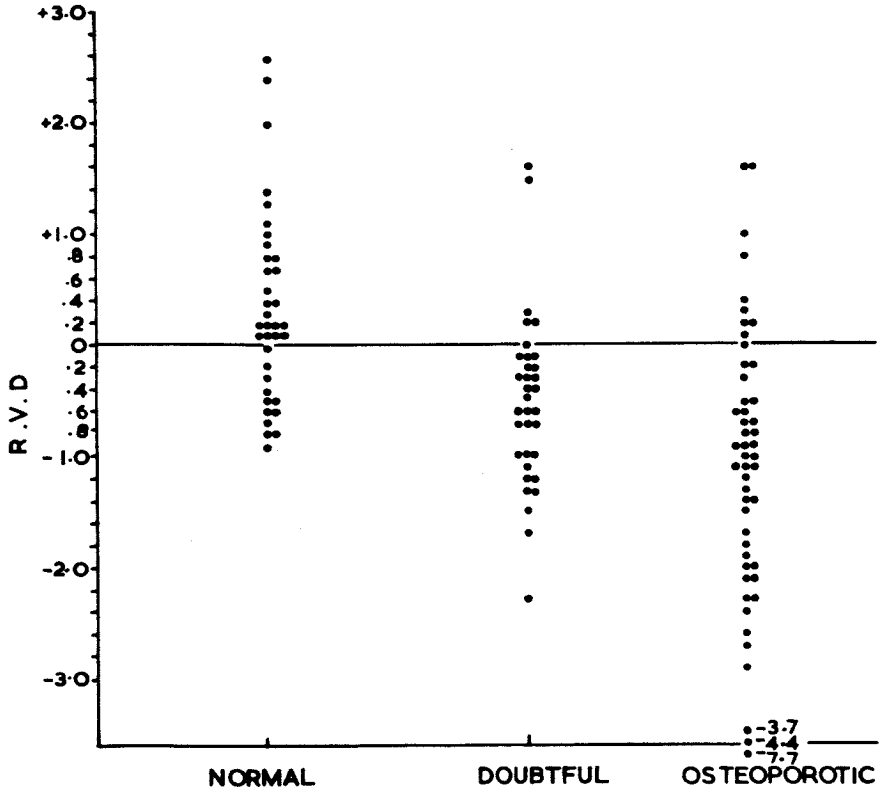

FIGURE 1.—Comparison between visual assessment of 119 unselected X-ray films with relative vertebral density. Visual assessment was performed without knowledge of the patient's age, sex, or R.V.D.

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"normal," "doubtful," or "osteoporotic," without knowledge of the patient's age, sex, or R.V.D. Films were classified as osteoporotic if there was vertical trabeculation, biconcavity, or compression present; they were classified as "normal" if none of these features were present and vertebral density appeared normal; they were classified as "doubtful" if there were any doubts about vertebral density. Comparison of this simple (and highly reproducible) classification with the R.V.D. (fig. 1.) shows reasonable agreement between the two. However, there are two anomalous films with high R.V.D. values which were classified as "osteoporotic" and two as "doubtful": one of these is a case of acute osteoporosis following an artificial menopause, one is a case of osteomalacia, and two are cases of renal failure.

In the osteoporotic case the anomaly consists of the association of biconcavity and compression (fig. 2) with normal density, suggesting that mechanical strength and X-ray density are not necessarily determined by the same factor. Repeated X-rays on this patient have confirmed this anomaly, which is emphasized by the fact that her metacarpal cortical thickness is normal (52%). Figure 3 illustrates this paradox again and shows that there is little relation betwen R.V.D. and the biconcavity index.

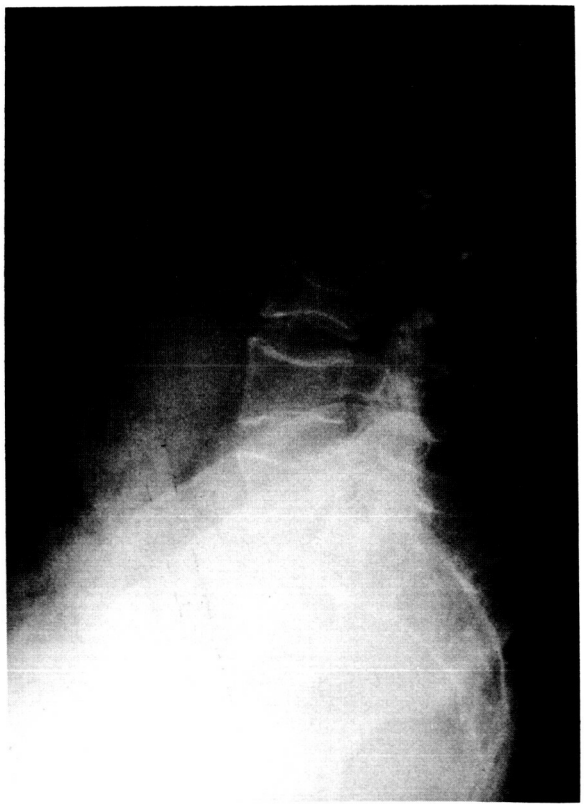

FIGURE 2.—Lumbar spine X-ray in a case of acute postmenopausal osteoporosis with normal vertebral density and normal metacarpal cortical thickness.

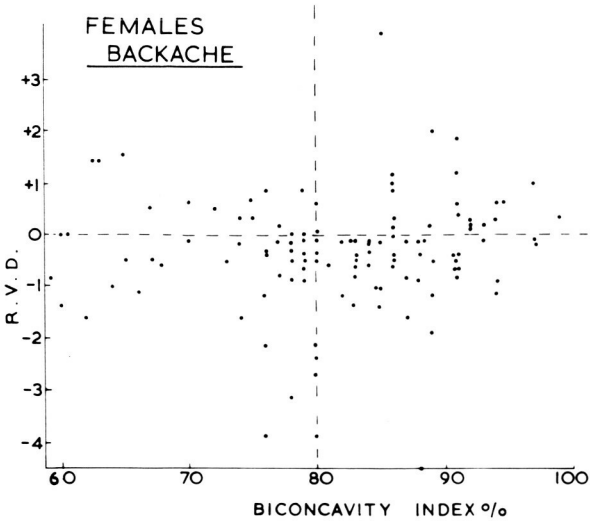

FIGURE 3.—Relation between biconcavity index and relative vertebral density in women with backache.

We have frequently observed an apparent sclerosis of the vertebrae in osteomalacia not unlike that which may be seen in renal failure (fig. 4), and we find that the R.V.D. tends to confirm this (figs. 8 and 9). While it is strictly true that bone density may be reduced *either* by reduced bone volume (osteoporosis) *or* by reduced bone mineralization (osteomalacia), it seems that, in the spine at least, the osteomalacic process tends to produce a different appearance from the osteoporotic one. Osteoporosis may be associated with osteomalacia, and this probably explains why an osteolacic spine is sometimes indistinguishable from an osteoporotic one. Cortical thickness tends to be low in the osteomalacia cases (figs. 8 and 9), but whether this is actually osteoporosis of the metacarpal or osteomalacia (or both), cannot be stated.

FIGURE 4.—Lateral lumbar spine X-ray in a case of osteomalacia to show apparent sclerosis of the vertebrae.

PATIENTS WITH BACKACHE

We have previously reported that patients referred with backache and a presumptive diagnosis of osteoporosis have X-ray indices rather lower than those of normal people, but they represent an extension of the normal state rather than a qualitatively different state. This supports our general concept that there is a continuum from normality into osteoporosis depending upon the amount of bone present. This is illustrated in figure 5 which shows how our original normal series appears to continue onward and downward into our backache series.

Much of the same is true of densitometry. We have described the fall of relative vertebral density (R.V.D.) with age in normal individuals elsewhere in this volume. Figure 6 shows how the patients with suspected osteoporosis extend from the normal range downward as though osteoporosis were no more than a con-

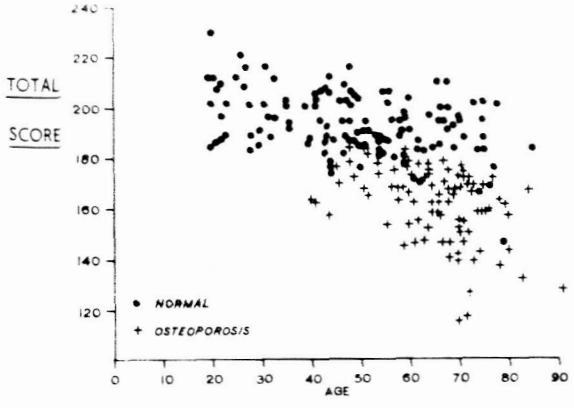

FIGURE 5.—Relation between total X-ray score (sum of metacarpal, vertebral, and spinal indices) and age in normal subjects and in cases of suspected osteoporosis to show how the former merge into the latter.

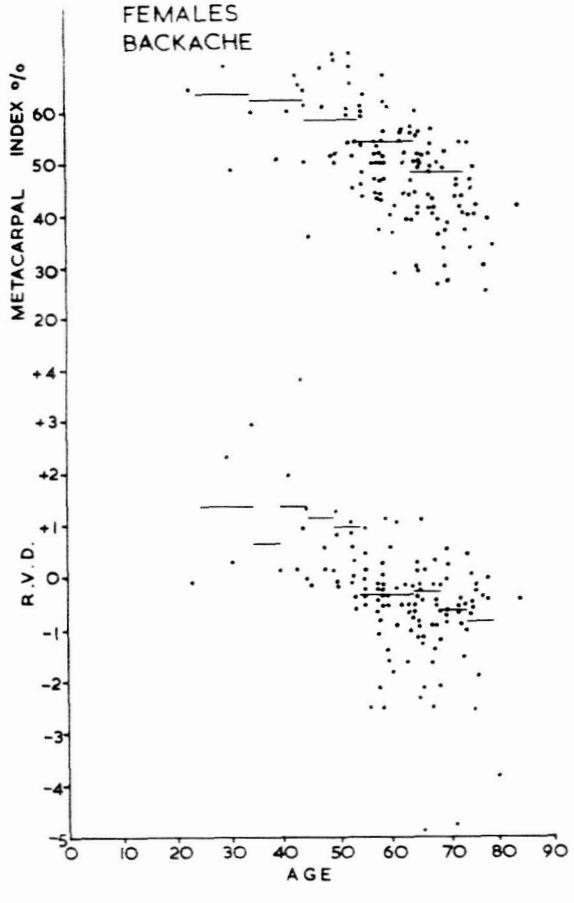

FIGURE 6.—Relative vertebral density (below) and metacarpal index (above) related to age in women presenting with backache. The horizontal bars represent the normal mean values.

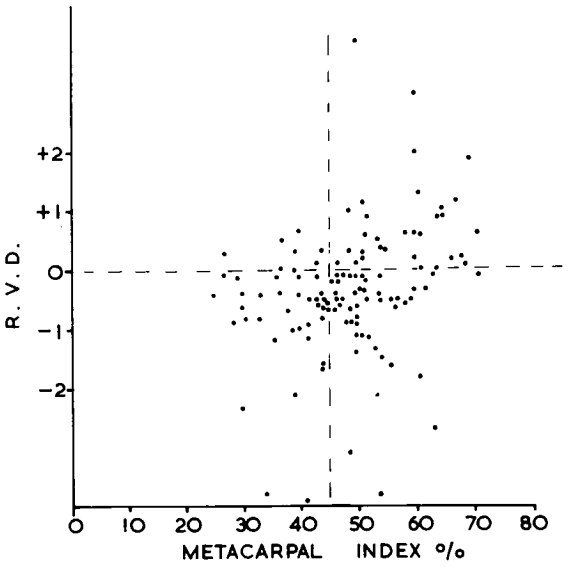

FIGURE 7.—Relation between relative vertebral density and metacarpal index in patients presenting with backache (compare with fig. 17).

tinuation or accentuation of a normal process. The relation between R.V.D. and metacarpal index is also very similar in the cases of suspected osteoporosis to what it is in the normal women, although both the R.V.D. and metacarpal values tend to be lower (fig. 7).

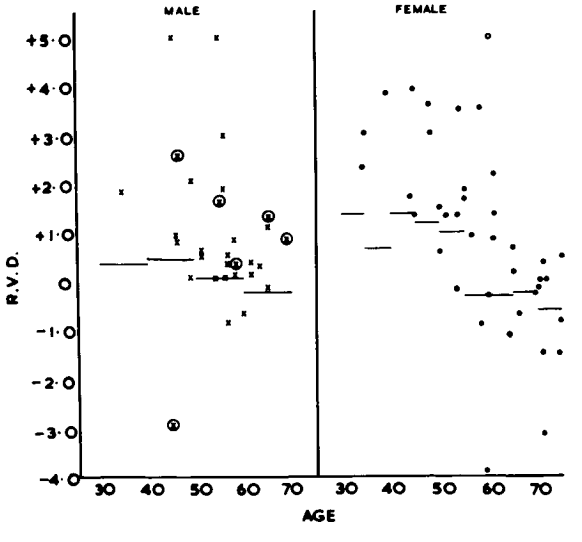

FIGURE 8.—Relative vertebral densities plotted against age in 30 men and 39 women with steatorrhoea. Horizontal bars represent mean normal values (× and ●=osteomalacia).

PATIENTS WITH STEATORRHOEA

R.V.D. values in 30 men and 39 women with steatorrhoea are shown in figure 8. There is a distinct tendency for the values to be high, especially in the mean and particularly in cases of osteomalacia. The corresponding metacarpal indices are shown in figure 9 and are distinctly low. Thus, the relation between compact and trabecular bone is abnormal in steatorrhoea with disproportionate loss of compact bone.

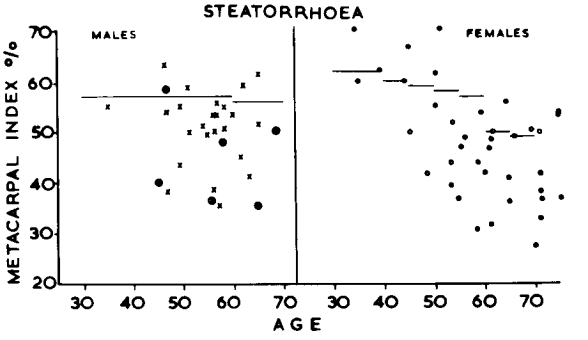

FIGURE 9.—Metacarpal indices plotted against age in 30 men and 39 women with steatorrhoea (× and ●=osteomalacia).

CORTICO-STEROID THERAPY

Assessment of vertebral density in patients on cortico-steroid therapy is immensely complicated by the number of variables involved. There is the age and sex of the patient, the underlying disease, the type of cortico-steroid used, and duration of therapy. Simple measurement of X-ray density or cortical thickness is, therefore, by itself of little value. We have tried to overcome this to some extent by converting all the cortico-steroids into their equivalent in grams of cortisone and examining the spinal density in relation to the total dosage given. This has yielded the results shown in figure 10 and suggests that the effect of cortico-steroids on spinal density is a function of the total dose given. The relationship shown in figure 10 cannot be explained by the age of the patients, which is not related to the total dose given.

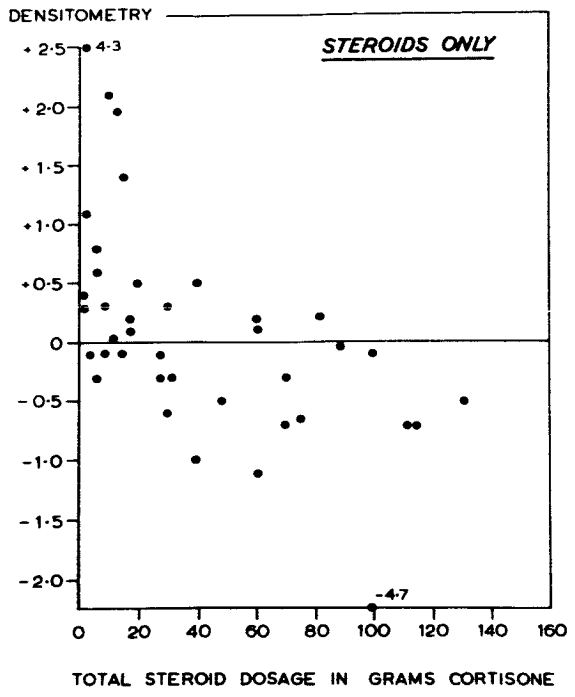

FIGURE 10.—Relation between relative vertebral density and total steroid dosage expressed as grams of cortisone in patients on cortico-steroid therapy.

EFFECT OF CALCIUM SUPPLEMENTS

Calcium glycerophosphate grams, 6 daily (1 g. Ca), have been given to 66 patients for over 1 years. Figure 11 shows the change in R.V.D. related to the subjective response of the patient. There does not appear to be any consistent change in spinal density on calcium therapy.

CONCLUSIONS

The validity of our procedure for measuring vertebral density appears to be supported by the following observations:

1. The relative vertebral density falls with age, especially in women after the menopause.
2. Relative vertebral density is generally related to metacarpal cortical thickness, at least in women.
3. There is general agreement between visual assessment of films and the relative vertebral density.

On the other hand, the precision, accuracy, and reproducibility of the method still leave a good deal to be desired; and the occasional normal values observed in patients with biconcavity require explanation. Finally, the effect of calcium therapy on spinal density is disappointing, but we do not know whether this is the fault of the therapy or of the densitometry.

COMMENTS

Dr. SMITH. In a study of two thousand women comparable to your normal group which we have surveyed in a similar fashion, we find that between the ages on 65 and 70 years 11% have fractures (one or more vertical compression fractures) and that 20% in the age group 70 to 75 have the same findings and are

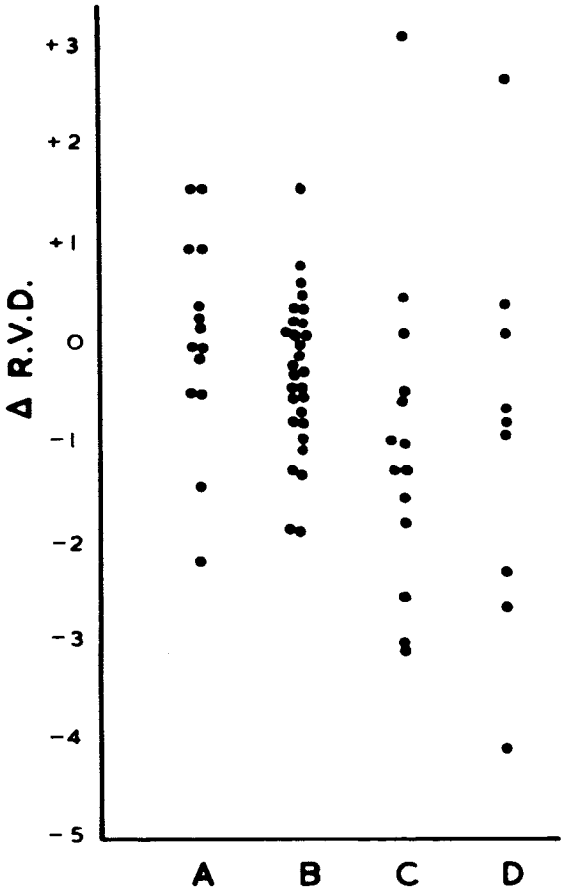

FIGURE 11.—Changes in relative vertebral density in 66 patients treated with calcium glycerophosphate supplements for at least 1 year. The cases have been classified according to subjective responses as follows: (a) pain free; (b) much improved; (c) doubtful improvement; (d) no change.

asymptomatic. I share your conviction that there is little if any relationship between the degree of osteoporosis with or without fractures and symptoms.

Dr. NORDIN. How did you select your two thousand?

Dr. SMITH. These were ambulatory outpatients who came in for routine physical examinations. I have a question in regard to the aorta and the spine. This seems rather peripheral, but I think it may be important ultimately. Did you correlate whether calcification was present or not, or did you quantitate it? I can see how osteoporosis might not correlate with whether aortic calcification is present or not, but did you give a score? We have done this with the two thousand patients and we find a highly significant correlation when you give aortic calcification, a score, age corrected.

Dr. NORDIN. No, we did not. It was in the British Journal of Radiology about 6 months ago. This was done on a lumbar basis. We did give a quantitative score but we did not use it. We thought it was too subjective. On an all or none basis, there was only one 10 year group in which there was even a suggestion of a correlation. I agree that in practice one sees the calcium in the aorta in association with osteoporosis; one feels there must be something in it.

Dr. SMITH. Osteophytosis or hypertropic changes do correlate inversely.

Dr. NORDIN. We hardly ever see them in osteoporosis. This must be of some interest, I agree.

Dr. URIST. We are doing a decade study. We are measuring the amount of calcium in milligrams per kilogram of aorta in 50 cases. I am now doing the aorta, fascia, skin, and lateral views of the dorsal and lumbar spine, and I am trying to quantitate this. My sample is much different from Smith's, Nordin's, Rich's, or Whedon's; they see a much younger age group. I do not see them until they have fractures. Out of a thousand cases I would only see the end picture of this spectrum.

Dr. RICH. I was very interested in the association between spines and fingers. This seems to bear out that many people feel that trabecular bone and cortical bone may react differently in these diseases. I can't comment on a thousand cases, but in one patient who was treated with fluoride for 2 years, we have seen X-ray density changes. Others have also seen these changes. I think Dr. Vose, in association with the group at Lahey, has seen them in a couple of patients. My purpose, however, is to indicate where the changes are seen. This is over a period of a hundred weeks of treatment. The changes in the main portion of phalanx, using Schraer's method interpreted by Dr. Schraer in terms of value changes, are not great, maybe 10%, which would probably be the same as the 10% change in the ratio that you measure, whereas the os calsis increased by twice that much.

Using Doyle's method, probably in our hands not as accurate as Schraer's method in his hands, the changes in the distal several centimeters of the ulna where there is trabecular bone were three or four times as great as in the area which is primarily cortical bone.

I want to make one comment on the sclerosis you see in osteomalacia, which is extremely interesting and to me seems a little parodoxical. Since there is a great deal of secondary hyperparathyroidism in such a patient, one wonders whether this sclerosis could somehow be related to increased turnover. I wonder if you had any vertebral report on the hyperparathyroidism patients. We think primarily they are sclerotic.

Dr. NORDIN. I had the same feeling that this could be a manifestation of hyperparathyroidism. In a vague sort of way it might be, inasmuch as you see it in renal failure. I agree with you. I do not have the figures on the hyperparathyroidism cases. I think there is one possible exception. When you come to histology, I am not sure what histological effect hyperparathyroidism would produce.

Dr. RICH. There is also increasing evidence that in addition parathyroid hormone does stimulate bone formation.

Dr. NORDIN. I agree.

Dr. GILSON. Dr. Cohen yesterday proposed a multi-gamma analysis method of determination of various components of the bone anatomy. I would like to expand upon this to show you where we have gone and where we have not gone, where we have bumped our heads, and where we have been successful.

In about 1962, we decided to make some naive attempts in the measuring of bone densitometry using radio isotope techniques. We tried this with simple densitometers and, of course, we did not get what we wanted. In defense of the bone densitometric people using film techniques, I must say we certainly were not doing the type of work which has been expounded here. I had the good fortune to fall upon a monograph by Dr. Carl Monnel, and his classic work stimulated us to think more about the monochromatic energies. The first attempt was to use Tm^{170}. At this time the search for what was available in monochromatic sources in the low energy range was pretty barren. I would like to show you first the spectrum of Tm^{170}. Tm^{170} has a gamma ray of approximately 53.1 and in the 80 region. We thought it would be just dandy because we decided on a multi-gamma technique. Using thin sources in small quantities we obtained very good spectra and very good absorption measurements on aluminum foils and other absorbers. As soon as we got into larger sources, unfortunately the high energy B-rays of thulium showed up and we had a spectrum that just about knocked it out as an isotopic choice. The spectrum, of course, was no longer monochromatic.

It was soon after this that Dr. Cameron recorded the use of I^{125}. We immediately obtained the source and went through his techniques and had absolutely no trouble in reproducing his results.

We decided to take a look at some of the bremsstrahlung sources and actually used and constructed this particular type device. It basically consists of a rather large Sr^{90} source upon which target materials are placed and a small X-ray generator at the source bombarding the various target materials and the shutter, giving us a flux of various "K" alpha radiation.

Now, we have relatively good clean curves. With a Cerium Uranium target, we obtained quite good peaks. We were again very happy except that our flux once again did not measure up to what we wanted. The size of the shielding material became so prohibitive as we went up in size that we had to abandon this. Now, why are we spending all this time looking for a multigamma approach? In Dr. Cameron's laboratory, if he gave me a result, I would be absolutely sure this was correct. In the reseach laboratory with many of the instruments devised by the investigators and with close attention to detail, the techniques work fine. The problem is that when this is turned into a clinical laboratory (and I am a radiologist), we know that it is important to try to eliminate as many of the variables as possible to make this a practical clinical tool. We wanted to take as many of the variables out as we could, and Dr. Cameron I am sure will tell me I have introduced more variables by adding another parameter. We feel, however, that, in the long run, probably in the clinical lab, not in the closely controlled research lab, but in the clinical lab, this will be less of a problem.

The problem is what to do about measurement of bone density in flight. I believe from what I can read and from what I have heard that measurements of actual soft tissue parts in flight would be a very cumbersome chore. I think over a long period of time we cannot assume that the soft tissue part is going to stay the same size. There will probably be changes in mass of soft tissue. If every time one has to do a measurement in flight one has to compress parts between various restraining tools, I think the procedure in flight, although it is a very simple one in theory, would become a very cumbersome chore.

The true gamma technique would obviate the necessity for these soft tissue measurements. This again is one of the reasons we feel, so far as NASA is concerned, that the two-time technique has possibilities. If one goes to the arm, the hand, or the os calsis, it is relatively easy to measure these parts and to restrain them between plexiglass sandwiches. In the head of the femur, for instance, or neck of the femur, I think this becomes more difficult. I am not arguing about the basic physical premise of the single gamma technique. I am simply saying that I think for many applications this will not be as satisfactory as using the multigamma technique which will obviate the necessity for the additional measurements.

Utilizing the bremsstrahlung inspection-mentioned sources, we obtained very good tracings. One could go out and trace over the soft tissue part to find out where the soft tissue started and where the cortical bone was and one could actually see the marrow cavity on the opposite side. We measured these cavities on several animals. The determination of bone size per se is quite accurate with a very highly confined collimated beam.

A machine has been constructed for the explicit purpose of using the multigamma technique. Right now we are most interested in just analyzing the dual-gamma measurement. We hope to go on to the tri-gamma measurement for analysis of the third factor and so on up to seven.

There is no one technique that will solve all the problems. Yet I think this is a technique which has to be investigated. So many of the problems that we have in nuclear medicine or in medicine in general involve the measurement of multicompartmental systems in dynamic states. Here we have a very good physical model in a sense. Things are not changing during the brief time that the exposure is made. We know a good many of the physical parameters involved, and for the purposes of the measurement everything is in steady state. I see no reason why one should stop with a one-gamma system when it is theoretically (and I use the word theoretically in context here) possible to go on to greater accuracies.

The machine uses a Siemen's tube from their electrometer apparatus. We closely control the ripple of our high voltage supply and try to keep the output stable. We hope to go on to better generators and more stable power supplies.

We use a vibrating electronic type ionization chamber, and have used GM counters and scintillation detectors. We found this electrometer very useful. You can produce a multiplicity among monochromatic X-ray beams utilizing proper filtration.

The reason for the development of the multigamma technique is that we have a problem that is in a stable state, where the basic laws of physics can be utilized. These mass absorption measurements of layers have been done since the very first days of radiation. I think it is possible, utilizing known absorption coefficients and known, carefully collimated monochromatic beams, to assay not just the bone content of an individual's part, but to measure the fat to muscle ratio, and the fat to lean mass ratio. Computer techniques are obviously going to be needed in the future.

We think this is a new approach which we feel has fertile ground. We are in the beginning of it.

Dr. CAMERON. Maybe Dr. Gilson feels I am against the multigamma technique. In fact, in *Science* we pointed out the formulas which are essentially identical

to the ones Dr. Cohen presented yesterday. You are absolutely right; in theory it is possible.

I think Dr. Gilson also is aware of the Symposium on Low Energy X-ray Sources, where we showed the use of a two-component system to determine the fat to muscle ratio in phantom. But again I want to repeat: as you go further, there are some very difficult technical problems. I think anybody who can lick them is going to have to work hard. I am willing to encourage them to do so, but I do want to caution them.

Estrogens and Postmenopausal Osteoporosis

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LAWRENCE H. LANZL
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It has long been known that with the cessation of ovarian function, or the decrease of ovarian function to a point below adequate amounts to maintain normal adult function, changes occur throughout the body. These changes occur at about the age 45 to 55 in the usual course of events. The same changes occur prior to the normal menopause in the case of bilateral oophorectomy performed at an age prior to the natural decrease or complete cessation of production of estrogens. As a woman approaches and passes through the menopause, there also occurs an alteration in the function of the adrenal cortex. The production of gonadal-like hormones, particularly estrogen, by the adrenal cortex diminishes steadily with aging. However, the production of glucocorticoid hormones by the adrenal cortex persists at the adult level. It can be said that, in general, the gonadal hormones such as estrogen have an anabolic action upon protein and osseous tissues. The glucocorticoid hormones have an antianabolic or, in some cases, a catabolic action upon the same tissues. In young adulthood there is a balance between the production of these two types of hormones. With aging, women develop a relative excess of the antianabolic steroids due to the continued decreased production of the gonadal hormones (from the ovaries and adrenal cortex) and the sustained production of the glucocorticoid hormones (from the adrenal cortex). Structures other than the skeleton also show changes following the menopause. These changes are chiefly atrophic. The target organs of the gonadal hormones, the vagina, uterus, vulva, and breasts, demonstrate profound atrophic changes. Other general physical changes also take place—thinning of the skin, loss of muscle mass, and changes in the hair. The addition of hormones such as stilbestrol can produce remarkable reversal of these changes. The addition of small amounts of stilbestrol causes thickening of the vaginal mucosa and changes in the glycogen content of the cells to premenopausal levels. The most important change, however, in the postmenopausal patient is the gradual loss of bone mass.

Osteoporosis, the loss of bone mass, is the most common systemic disease of the skeleton and, in its severe form, is the most disabling of all the degenerative (aging) effects of the menopause. In some patients this is mild; but in others it is severe and leads to complications such as bone pain, loss of height due to collapse of vertebrae, and the thinning of the long bones, which makes the patient more susceptible to fractures, particularly of the hip. The majority of patients with fractures of the hip in the postmenopausal female show osteoporosis, which is more severe than the so-called normal osteoporosis or decrease in bone mass, which is the finding in all patients who reach old age. In the group of postmenopausal patients, in whom the reduction of bone mass is accelerated,

clinical symptoms are usually present. Osteoporosis of a clinical degree is four to six times more common in females than males. Patients with postmenopausal and senile osteoporosis are improved by the addition of estrogens alone. The bone pain disappears and the decrease in height due to the collapse of vertebrae ceases. Many attempts to document the changes in the skeleton secondary to the addition of hormones have been made. Drs. Paul C. Hodges and Franklin C. McLean, working with Dr. M. Edward Davis, made radiographs on a large group of patients on supplemental postmenopausal hormone therapy, but were totally unable to detect any changes. In view of the material presented at this conference, this is not surprising.

Dr. M. Edward Davis, Chairman of the Department of Obstetrics and Gynecology at The University of Chicago, 25 years ago began giving stilbestrol as a replacement of natural (ovarian) hormones following oophorectomy. These patients with few exceptions have been on stilbestrol 0.5 mg, 3 times a week, continuously since oophorectomy. It was largely to study this group of patients that we invested time and effort in our I^{125} densitometry apparatus.

This group of patients has been studied extensively with serial EKG tracings, vaginal smears and biopsies, and serum cholesterol and lipid determinations to assess the effects of stilbestrol in retarding the aging process. The results of this study will be published shortly. The subjective feeling of well being in patients on stilbestrol postmenopause is well known. We are interested in objective evidence of retardation of aging; in our case, the possible effect of hormones on osteoporosis. We will present today the data on our studies of bone density in this group of patients.

We have compared the patients postbilateral oophorectomy with supplemental hormones (stilbestrol 0.5 mg, 3 times a week) against postmenopausal (natural or oophorectomized) controls without exogenous hormones, and a group of patients postmenopausal (natural or surgical) who for various reasons have had intermittent hormone supplementation. These patients were usually with oophorectomy done

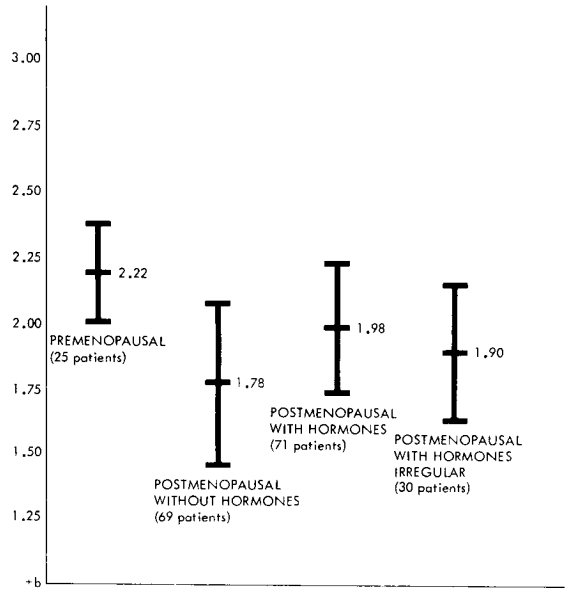

FIGURE 1.—Bone-density evaluation from January 17, 1964 to January 27, 1965, expressed as linear coefficients of absorption.

at other institutions who, after an interval of time postoophorectomy, have been placed on stilbestrol or, in a few instances, patients in whom stilbestrol has been discontinued.

The results of this study are shown in table I, which gives the linear coefficients of absorption for cortical bone (μ_{cb}) and over all bone

TABLE I.—*Bone-Density Evaluations (From Jan. 17, 1964 to Jan. 27, 1965)*

Group 1—Premenopausal normal females (25 patients).	$\mu_{cb}=2.83\pm0.24$ cm^{-1} $\mu_{tb}=2.22\pm0.19$ cm^{-1}
Group 2—Postmenopausal females without hormones (69 patients).	$\mu_{cb}=2.64\pm0.33$ cm^{-1} $\mu_{tb}=1.78\pm0.33$ cm^{-1}
Group 3—Postmenopausal females with hormones (71 patients).	$\mu_{cb}=2.75\pm0.20$ cm^{-1} $\mu_{tb}=1.98\pm0.26$ cm^{-1}
Group 2-3—Postmenopausal females with hormones for irregular time periods (30 patients).	$\mu_{cb}=2.70\pm0.22$ cm^{-1} $\mu_{tb}=1.90\pm0.27$ cm^{-1}

(μ_{tb}). The linear coefficient of absorption for postmenopausal (oophorectomized patients) is 1.98 ± 0.26 cm^{-1} as compared to 1.78 ± 0.33 cm^{-1} for the control group (patients postoophorectomy or postmenopausal without added hormones). The group of patients with intermittent supplemental hormones was 1.90 ± 0.27 cm^{-1}. The standard deviation given is that of the individual of the group and does not indicate the standard deviation of the mean. As one would anticipate, there is a wide range of values in each group. These patients were not selected because they were clinically osteoporotic but were placed on stilbestrol prophylactically. The data is shown here on the bar graph (fig. 1) and includes all the patients. Table II gives the linear coefficients of absorption of the patients grouped in 5-year intervals postmenopause. The standard deviation of an individual in each group is given and also the standard error of the mean. The data are depicted in this line graph (fig. 2) in which the patients are grouped according to years postmenopause.

There is, with aging, a gradual decrease in the linear coefficient of absorption in the controls without harmone supplementation. Here the patients have been grouped into 5-year increments postmenopause and are compared with the patients on stilbestrol. These patients with the additional hormone also have a decrease in the linear coefficient of absorption with increasing years postmenopause, but not to the same degree as the controls, and the close correlation of the two groups, zero to 5 years postmenopausal, is striking. The data on part of these patients are shown on this scatter graph (fig. 3) where, in addition, the patients with stilbestrol and without are compared to a group of premenopausal females where the linear coefficient of absorption (μ_{tb}) is 2.22 ± 0.19 cm^{-1}. There is some overlap in the groups as one would anticipate, but the difference between the three groups is evident.

The linear coefficient of cortical bone (μ_{cb}) of this same group of patients on semilog scale (fig. 4) results in a straight line. The measurement of the cortical bone thickness is not as accurate as the measurement of total bone. Measurements of bone thickness were made with a magnifying lens with a built-in scale.

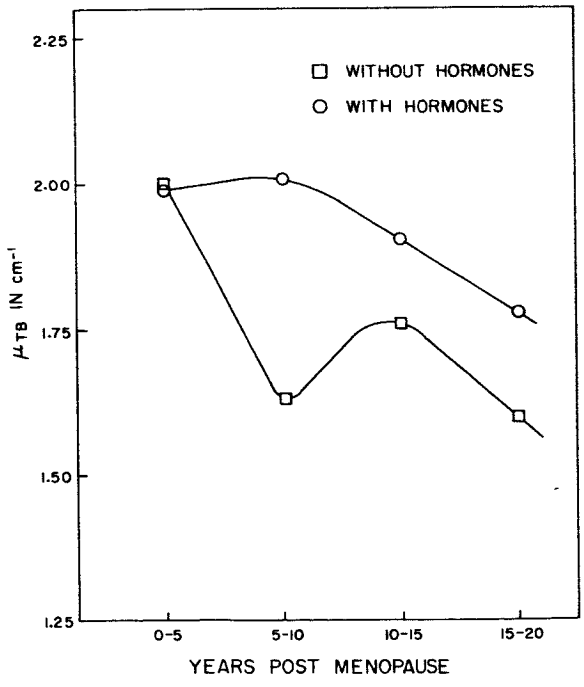

FIGURE 2.—Absorption coefficient versus years postmenopause.

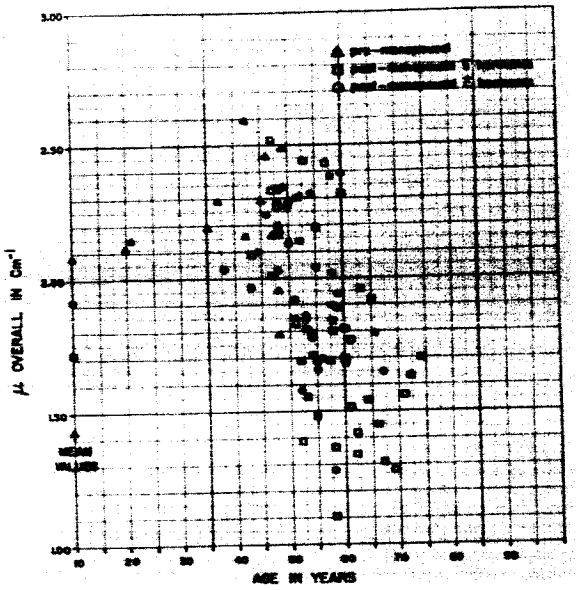

FIGURE 3.—Overall bone density coefficients versus age of patient, from December, 1963 to July 16, 1964.

TABLE II.—*Bone-Density Data Summary*

(January 17, 1964 through January 27, 1965)

Group	Number of patients	μ_{tb}	(Std. dev.) σ	(Std. error of mean) σ_μ	μ_{cb}	σ	σ_μ
△ 1	25	2. 22	±0. 19	±0. 04	2. 83	±0. 24	±0. 05
□ [2] 0–20	52	1. 79	± . 33	± . 05	2. 66	± . 32	± . 04
[2] 0–5	15	2. 00	± . 32	± . 08	2. 73	± . 24	± . 06
[2] 5–10	16	1. 68	± . 35	± . 09	2. 53	± . 30	± . 07
[2] 10–15	15	1. 76	± . 31	± . 08	2. 78	± . 34	± . 09
[2] 15–20	6	1. 65	± . 16	± . 06	2. 53	± . 41	± . 16
○ [3] 0–0	72	1. 98	± . 26	± . 03	2. 74	± . 20	± . 02
[3] 0–5	18	1. 99	± . 18	± . 04	2. 75	± . 22	± . 05
[3] 5–10	32	2. 03	± . 30	± . 05	2. 72	± . 24	± . 04
[3] 10–15	18	1. 91	± . 24	± . 06	2. 74	± . 19	± . 04
[4] 15–20	4	1. 78	± . 37	± . 18	2. 86	± . 21	± . 11

All μ and σ in cm^{-1}.
Subnumbers refer to number of years since menopause.

To test for the equality of two distributions of patients, we have used the two-sample test with unequal variances as developed by Welch (Brownlee, 1960). In this test we assume we have two independent samples from normal distributions with means and sampled variances.

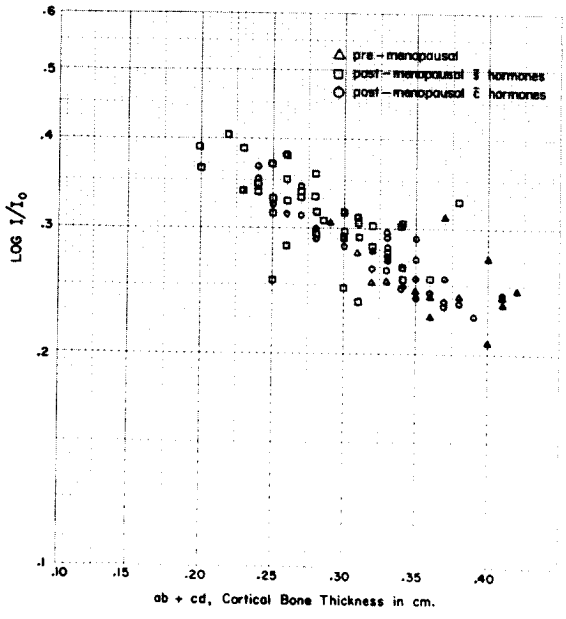

FIGURE 4.—Log I/I₀ versus cortical bone size, from December 1963 to July 16, 1964.

We wish to test the null hypothesis that these means are equal. From figure 2 we see that the absorption coefficient for the cases with and without exogenous hormone for the 0–5-year postmenopausal group are essentially the same. However, for the 5–10-year postmenopausal group, the odds against the two mean values being the same are greater than 99 to 1. For the 10–15- and the 15–20-year groups, the statistical differences are far smaller. The number of patients in the 15–20-year postmenopausal group are quite small, and it may not be possible to evaluate further this group at this time.

CONCLUSION

The results of our study indicate that:

1. There is a gradual loss of bone mass with increasing age (a decrease in the linear coefficient of absorption of total bone).

2. The linear coefficient of absorption of cortical bone is relatively unchanged. This supports the concept that osteoporotic bone is of the same composition as normal bone.

3. Comparison of three groups of patients postmenopausal demonstrates retardation of aging (osteoporosis) in patients with exogenous estrogens.

REFERENCE

BROWNLEE, K. A.: Statistical Theory and Methodology in Science and Engineering. John Wiley and Sons, New York, 1960, 235 pp.

COMMENTS

Dr. LANZL. Dr. Strandjord has shown the standard deviation assuming a normal distribution of those individuals. You notice some overlapping in these groups. How do you treat these data? We have taken two postmenopausal situations with and without hormones and broken them down into 5 year intervals. These two are quite close together for the first 5 years after menopause. Is there a meaningful difference thereafter? We have calculated the standard deviation of the mean and then evaluated the significance of mean, but by the test of Welch. This is a two-sample test with unequal variance. In Welch's test one asks the null hypothesis. If you were to repeat this experiment with this number, what is the probability that these two would be the same? It turns out, according to this test, that the odds are about 99 to 100 that you would not get these two means to overlap. Therefore, we conclude that this is a meaningful difference.

Dr. Strandjord asked me to summarize this last paper. First, there is a gradual loss of bone mass with increasing age, a decrease in the linear coefficient absorption of total bone. Second, the linear coefficient absorption of cortical bone is relatively unchanged. Third, comparison of three groups of postmenopausal patients demonstrates retardation of aging (osteoporosis) in the patients with exogenous estrogens.

Dr. NORDIN. Postmenopausal osteoporosis, we would all agree, is probably a phenomenon of trabecular bone. It is something that happens at the end of the fall with age. The compact bone is much less significantly associated with the menopause than the trabecular bone.

Dr. HURXTHAL. In 107 postmenopausal women out of a series of 400 random controls, we found no difference from the mean. Furthermore, in 11 castrates of over 30 years' duration we still found no difference.

Dr. NORDIN. In what bone?

Dr. HURXTHAL. In L–3. Also, in 50 patients with osteoporosis, that is, patients with symptomatic osteoporosis or patients exhibiting a marked decrease in density by X-ray, the density from zero to 4 years after the menopause was exactly the same as the density from 30 years after. I figured that the people who had low densities 4 years after the menopause had the low density before the menopause began. Therefore, the "postmenopausal" osteoporosis was present before the menopause. I agree with Dr. Arnold that under this theory you get in the end what you start out with.

Calcium Loss Studies During Human Bed Rest: A Preliminary Report

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Nelda Childers Stark Laboratory
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During the past 2 years, our team of research workers at the Texas Woman's University has been conducting an investigation on the effects of bed rest immobilization on metabolic and skeletal changes in healthy adult males. Seventeen men, screened for physical and psychological responses to batteries of examinations and tests, have cooperated in this investigation, which includes metabolic studies and studies of bone mass changes resulting from continuous bed rest for designated periods of time. This is a brief, preliminary report.

Five 14-day units of the investigation and one 30-day unit have been completed, although all data have not been analyzed. One 30-day bed rest unit currently is in progress. Each bed rest period has been preceded by an ambulatory preconditioning period and has been followed by an ambulatory period of reconditioning. The preconditioning, the bed rest, and the reconditioning periods combined are designated as constituting one bed rest unit. When an ambulatory period is investigated for comparison with a bed rest period involving the same level of calcium, preconditioning and reconditioning periods also precede and follow the ambulatory period under consideration.

SCREENING THE SUBJECTS

The subjects are given preliminary interviews by the director of the laboratory; and if they show no apparent undesirable characteristics or situations which might interfere with their participation in the project, they are sent to the Medical-Surgical Clinic in Denton where they are examined by two internists on the clinic staff who also are Visiting Professors at the Nelda Childers Stark Laboratories of the Texas Woman's University. The preliminary examinations consist of the following:

1. A thorough medical examination which includes a review of: eye, ear, nose, and throat systems; chest and gastrointestinal systems; and genitourinary systems, as well as a full examination of thyroid, heart (with an accompanying electrocardiogram), lungs, abdomen, genitalia, rectum, prostate, and reflexes.
2. Chest X-ray of the lungs and heart, diagnosed by the Visiting Radiologist on the TWU research staff.
3. Clinical laboratory tests which include the complete hematological series; the complete fasting blood sugar series; and a urinalysis series.

In order to secure information about their probable behavior during their participation in the study, the subjects also are given a neuropsychiatric examination at the clinic and psychological tests at the Texas Woman's University.

GENERAL PLAN OF THE INVESTIGATION

The men are kept at bed rest continuously for designated periods of time on planned diets, with calcium the chief dietary variable. While

they are ambulatory, they are assigned sedentary tasks for 8 hours daily in the various laboratories of the TWU Research Institute, with their hours of sleep and other activities regulated as nearly as possible.

While at bed rest, the men use only one pillow, and they remain recumbent throughout this phase of the study. They are bathed in bed, with their teeth brushed by trained male orderlies. All hygienic needs are cared for by the orderlies. They are spoon-fed by the dietitians on the project.

They pass the time by watching their respective hospital television sets equipped with ear phones and by reading through glasses equipped with prismatic lenses. Arm motion in the men is not restricted.

EXPERIMENTAL DIETS

The daily levels of calcium fed at the different 14-day periods have included 0.5 gram, 0.7 gram, 1.0 gram, 1.5 grams, and 2.0 grams, respectively. The 30-day bed rest unit, which has been completed, provided 2.0 grams of calcium per day, while the 30-day unit currently in progress provides 1 gram daily. The calcium levels have not been maintained in the order given here. Before any one bed rest is begun, the men are equilibrated with respect to bone mass of the central os calcis section on the diet which contains 1.5 grams of calcium per day, unless they are going on to a unit which includes 2.0 grams of calcium daily, in which case this calcium level is maintained throughout the unit.

The phosphorus in each unit is provided so that the Ca : P ratio falls within the 2 : 1 to 1 : 2 range suggested by Cantarow and Shepartz (1962), while not interfering with the protein provision. The remainder of the diet is held as closely as possible throughout the study to the following: 2400 calories, 90–100 grams of protein, 18–20 milligrams of iron, 10,000 International Units of vitamin A equivalency (including preformed vitamin A and provitamin A), 400 International Units of vitamin D, 100–150 milligrams of ascorbic acid, and minima of 1.6 milligrams of thiamine, 1.8 milligrams of riboflavin, and 21 milligrams of niacin per day. A minimum of 250 milligrams of magnesium is supplied daily, in line with the suggestion by Wohl and Goodhart (1960).

The diets are designed with respect to the types of foods selected and their preparation so as to fall in the low residue category. New cooperators in the study are informed about the effects of the low residue diet before they begin so that they will not worry about their probable infrequency of defecation.

Management of the diets is in the hands of a head dietitian who is certified by the American Dietetics Association and who is assisted by two other experienced dietitians. When the series of studies began, a basic diet was prepared which can be altered to accommodate the different levels of calcium. For each calcium level, three master menus have been prepared which are rotated and which allow for limited substitutions within foods of the same type in order to avoid monotony.

At each meal the same quantity of the solid and liquid foods are given to each subject, with the solid foods weighed on a calibrated gram balance and the liquids measured in a calibrated volumetric container. The subjects are encouraged to consume all foods but are not forced to do so. Rejected food is weighed and the results entered into the subject's record.

Whether the subjects are ambulatory or at bed rest, they are given 8 to 10 eight-ounce portions of water or of nonmilk liquids on a schedule throughout the day and early evening.

The calcium and phosphorus content of each food in each meal is determined by laboratory analysis, and the provisions of other nutrients are calculated from standard food composition tables by a program established in the TWU Data Processing Center.

TESTS MADE ON EXPERIMENTAL SUBJECTS

Table I outlines the units of the bed rest immobilization study to date and the subjects participating in each subdivision. It will be noted that three men have taken part in three units, four men in two units, and the remainder in one unit of the series to date. Some of those in

TABLE I.—*Units of the Bed Rest and Corresponding Ambulatory Phases of the Investigation*

Unit	Level of daily calcium provided	Length of the bed rest	Participating subjects in bed rest and corresponding ambulatory periods
1	1.5 grams	14 days	Subjects A, B, D, E.
2	0.7 gram	14 days	Subjects A, B, D, E.
3	2.0 grams	14 days	Subjects A, D, G, H.
4	0.5 gram	14 days	Subjects G, H, J, K, L.
5	2.0 grams	30 days	Subjects G, L, M, N, O.
6 and 7	1.0 gram		Subjects P, Q, R, S, T.
	Begin as 14-day unit, but continued without change of any kind as a 30-day unit		

Units 5, 6, and 7 will participate in a forthcoming unit. Table II gives the tests made on the subjects in each unit and the frequency with which they are administered.

Cardiovascular tests have been added by the National Aeronautics and Space Administration to the 30-day bed rest unit now in progress as well as to the 30-day ambulatory period which preceded and to the 30-day ambulatory period which will follow this bed rest period.

PRELIMINARY REPORT ON FINDINGS

Calcium Balance

In spite of metabolic differences, all subjects were in calcium balance during the equilibration period during which they were fed 1.5 or 2.0 grams of calcium daily, depending upon the level of calcium to be fed during bed rest. On the other hand, all subjects were in negative balance to a minor or major degree during all

TABLE II.—*Tests Administered to the Subjects During the Specified Units of the Investigation*

Unit	Test	Frequency of test
1, 2, and 3	Blood tests for hematology; plasma ascorbic acid, vitamin A, carotene, and phosphorus; serum calcium, acid and alkaline phosphatase, total protein, albumin, and cholesterol; and urinary tests for thiamine, riboflavin, and N'Methylnicotinamide.	Weekly.
	Urinary and fecal tests for calcium and phosphorus	Daily.
	Roentgenograms of lateral os calcis	Daily during bed rest, daily or every other day during ambulation.
	Roentgenograms of the hand with emphasis on phalanx 5–2.	Weekly.
4	Tests the same as for Units 1 to 3 except that a lateral view of the patella was added.	Weekly.
5	Tests the same as for 4 except that urinary tests for nitrogen, creatine, and creatinine were added.	Daily.
6 and 7	Tests the same as for 5 except that urinary tests for 17 keto-steroids and 17 cortico-steroids were added.	Daily.

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TABLE III.—*Data on Calcium Consumption, Calcium Excretion, and Calcium Balance During Respective Bed Rest Periods*

Bed rest unit	Mean calcium planned daily (grams)	Mean calcium consumed daily (grams)	Mean calcium excreted daily (grams)			Mean calcium balance (grams)	Data for ambulatory control period for comparison (grams)
			Urinary	Fecal	Total		
(1) 14-day:							
Subject A	1. 500	1. 457	0. 424	1. 232	1. 656	−0. 199	[a] +0. 394
Subject B		1. 316	0. 406	1. 065	1. 472	−0. 156	+0. 008
Subject D		1. 526	0. 354	1. 277	1. 631	−0. 105	−0. 018
Subject E		1. 484	0. 190	1. 529	1. 719	−0. 235	+0. 196
(2) 14-day:							
Subject A	0. 700	0. 659	0. 411	0. 602	1. 013	−0. 354	[a] +0. 111
Subject B		0. 636	0. 425	0. 541	0. 966	−0. 330	+0. 021
Subject D		0. 686	0. 318	0. 653	0. 971	−0. 285	+0. 095
Subject E		0. 675	0. 135	0. 824	0. 959	−0. 284	+0. 133
(3) 14-day:							
Subject A	2. 000	2. 017	0. 460	1. 814	2. 274	−0. 257	[b] +0. 430
Subject D		2. 038	0. 380	1. 978	2. 358	−0. 320	+0. 425
Subject G		2. 084	0. 406	1. 971	2. 377	−0. 293	+0. 260
Subject H		1. 910	0. 325	1. 790	2. 115	−0. 205	+0. 182
(4) 14 day:							
Subject G	0. 500	0. 466	0. 260	0. 722	0. 982	−0. 516	[a] +0. 376
Subject H		0. 431	0. 257	0. 801	1. 058	−0. 627	+0. 221
Subject J		0. 395	0. 245	0. 672	0. 917	−0. 522	+0. 122
Subject K		0. 431	0. 212	0. 726	0. 928	−0. 507	+0. 249
Subject L		0. 436	0. 178	1. 071	1. 249	−0. 813	+0. 145
(5) 30-day:							
Subject G	2. 000	1. 876	0. 451	1. 632	2. 083	−0. 207	[b] +0. 464
Subject L		2. 033	0. 322	1. 868	2. 190	−0. 157	+0. 353
Subject M		1. 896	0. 326	1. 717	2. 040	−0. 144	+0. 546
Subject N		2. 062	0. 478	1. 694	2 172	−0. 110	+0. 263
Subject O		2. 192	0. 422	1. 976	2. 398	−0. 206	+0. 287
(6) 14-day	1. 000	Data not yet analyzed.					
(7) 30-day	1. 000	This unit of the study not yet completed.					

[a] Calcium intake level 1.5 grams/day during control ambulation period.
[b] Calcium intake level 2.0 grams/day during ambulation period.

bed rest periods, as determined by a comparison of the intake and outgo of this element, the latter in urine and feces. The calcium tests were made in triplicate by trained biochemists.

Table III summarizes the data on calcium consumption, calcium excretion, and calcium balance for the bed rest immobilization units of the investigation for which the analyses have been completed, together with the balance status when the subjects were ambulatory on the equilibration levels of calcium intake. All balance data in the table are given as average grams per day during the period specified.

The table shows that the degree of negative balance varied between subjects for each bed rest level of dietary calcium, although there was a general relationship between the level of calcium intake and the extent of the negative

balance. The feces are shown to change somewhat more widely than the urine in calcium excretion at different levels of calcium intake.

The highest degree of negative calcium balance appeared in the 14-day bed rest period during which only 0.5 gram of calcium was provided, with somewhat less than this amount of calcium consumed. Rejection of bread with its content of dried milk powder was a major reason for the lowered calcium intake of this group.

Calcium Consumption and Excretion Data in Earlier Investigations

Of major investigations in which intake and outgo of calcium are reported, the following may be cited.

In 1945, Howard et al. of the Johns Hopkins Hospital investigated the effect of prolonged bed rest on 13 male subjects, varying in ages from 14 to 64 years, who had been hospitalized because of femoral or tibial fractures. Four other patients were studied before and after femoral osteotomy. All were in a good state of nutrition, and none manifested any abnormalities except those for which they were hospitalized. The patients were immobilized in extensive casts. Analyses of urinary calcium, phosphorus, and nitrogen were made as well as various blood tests.

After immobilization, urinary calcium excretion rose steadily on constant diets as did the quantity of phosphorus in the urine. The amount of urinary calcium excreted on dietary intakes of 2.0 grams per day ranged from 495 to 600 milligrams. Values for calcium in the feces were not reported, and hence calcium balance status could not be calculated.

Two reports were issued in 1948 and 1949 from the Department of Medicine of Cornell Medical College, the New York Hospital, and the Russell Sage Institute of Pathology on the results of immobilizing healthy subjects in terms of various metabolic and physiologic functions. The first of these, reported by Deitrick et al. (1948), involved four healthy young men (who participated in a preliminary control period of 6 to 8 weeks, an immobilization period of 6 weeks for one pair and 7 for the other, and a recovery period of 4 weeks for one and 6 for the other. During the immobilization period the men were placed in bivalved casts extending from the umbilicus to the toes. They remained in these casts constantly throughout the immobilization period except for the use of the bedpan and for ergometer and tilt table tests. The ergometer and tilt tests were performed periodically during the immobilization period. The time during which the men were free of the casts averaged 30 to 40 minutes daily.

Immobilization brought about a prompt increase both in urinary and fecal calcium, which continued and reached a maximal peak at 4 to 6 weeks. Whedon (1960) later called attention to the similarity of the pattern of calcium excretion in this study with that seen by Howard et al. (1943), cited above. Calcium excretion (urinary and fecal) recovered slowly and continued to show values greater than the control period for 3 weeks.

The second study made by these investigators was reported by Whedon et al. (1949). They were concerned with measures which might improve the disadvantageous outcomes of long immobilization. As one means of reducing the ill effects of long confinement to bed, the Sanders slowly oscillating bed was investigated with three normal healthy young men who had taken part in the previous immobilization study with fixed beds. Thus the metabolic and physiologic data from the former study could serve as a control for this study.

Urinary calcium in this study increased less rapidly than in the fixed-bed experiment. For all three subjects the maximum urinary excretion for a 3- to 4-day pooled specimen ranged from 155 to 458 mg. per day, with an average maximum of 311 mg. This is to be contrasted with the maximum urinary calcium excretion of the same three subjects in the fixed-bed experiment of 140 to 594 mg., with an average maximum of 362 mg. During the recovery phase following immobilization in the oscillating bed, calcium excretion decreased toward control levels more rapdily than it did in the fixed-bed study.

In a study by Whedon and Shorr (1957), 11 subjects were studied who were the victims

of the seriously immobilizing disease, acute anterior poliomyelitis. Maximal negative calcium balance occurred on the average of 9 weeks after onset and ranged from −0.420 to −0.683 gram per day among seven patients, with a mean of −0.540 gram per day. Negative calcium balance continued for an average of 7 months, with positive balance regained approximately after patients began to stand on their feet. Phosphorus excretion, on the other hand, increased very promptly, in contrast with calcium excretion, with the increase entirely in the urinary sample. Maximal phosphorus in the urine ranged between 1.07 and 2.13 grams per day, based on a 3-day average. Return to a positive phosphorus balance on diets providing from 1.35 to 1.66 grams daily varied from 8 to 9½ months after onset of the disease.

A study of cardiodynamic and metabolic effects of prolonged bed rest conducted at Lankenau Hospital, Philadelphia, was published by Birkhead et al. (1963). The study was sponsored by the 6570th Aerospace Medical Research Laboratories, Wright-Patterson Air Force Base, Ohio.

In this investigation, four healthy men served as the subjects of the study. The men were admitted to the Metabolic Ward of the hospital 3 weeks before the beginning of the bed rest period, with the first part of the preliminary period concerned with initial examinations and tests, and the last 18 days devoted to physical training. The bed rest period covered 42 days, followed by an 18-day retraining period. During the bed rest phase of the study, the only activity allowed the men was that of arms and legs.

During the study the subjects were on a weighed diet of 2523 calories—77 grams of protein, 74 grams of fat, 385 grams of carbohydrate, and 1.724 grams of calcium. Analyses of the diet for calcium and nitrogen were made at frequent intervals. The diet consisted primarily of a liquid formula.

The most marked change found during bed rest included definite increases in excretion of calcium and phosphorus. In some cases the urinary excretion of calcium more than doubled, and the increase began during the first 6 days in bed.

In a bed rest study conducted at the Texas Institute of Rehabilitation and Research, Texas Medical Center, Houston, reported by Vogt et al. (1965) to the National Aeronautics and Space Administration, which had sponsored the project, results of bone mass measurements as determined by radiographic densitometry and calcium and phosphorus balance trials were reported. According to the experimental design of the study, six healthy young men, 21 to 34 years old, participated in two 14-day bed rest periods which differed only in the provision of controlled isometric exercise during the second period.

After the bed rest period began, increases in calcium and phosphorous losses, both in urine and feces, were experienced. With a diet which provided approximately 1.0 gram of calcuim per day, mean negative calcium balance levels were found for all of the participants. When the same men engaged in a second bed rest period during which controlled isometric exercise was taken at defined times per day, the output of calcium tended to be somewhat lower than was the case when the men were at bed rest without the exercise. Moreover, the determinations which were made on these men by the TWU method of radiographic densitometry showed greater conservation of bone during the bed rest period when isometric exercise was taken in bed than during the previous bed rest period when no exercise was taken.

Distribution of Calcium Between Urine and Feces

This investigation has shown to this point that the percentage of calcium excreted in the feces is far higher than that excreted in the urine, which is consistent with the findings of other investigators. This is contrary to the situation with phosphorus, which is excreted in far larger percentages in urine than in feces.

Although the distribution of calcium between feces and urine was found to vary with individual men, the percentages of calcium in the urine in these studies have been highest when

TABLE IV—*Per Cent Distribution of Excreted Calcium Between Urine and Feces*

PART A. EXCRETION LEVELS DURING BED REST

Subjects	Percent of excreted calcium					
	Bed rest 2 on lowest dietary calcium level (0.7 gram/day)		Bed rest 1 on intermediate dietary calcium level (1.5 grams/day)		Bed rest 3 on highest dietary calcium level (2.0 grams/day)	
	Urine	Feces	Urine	Feces	Urine	Feces
Subject A	40. 6	59. 4	25. 6	74. 4	20. 2	79. 8
Subject B	44. 0	56. 0	27. 6	72. 4	X	X
Subject D	32. 8	67. 2	21. 7	78. 3	16. 1	83. 9
Subject E	14. 1	85. 9	11. 1	88. 9	X	X

PART B. EXCRETION LEVELS DURING AMBULATION

Subjects	Percent of excreted calcium					
	Ambulatory period on lowest dietary calcium level (0.7 gram/day)		Ambulatory period on intermediate dietary calcium level (1.5 grams/day)		Ambulatory period on highest dietary calcium level (2.0 grams/day)	
	Urine	Feces	Urine	Feces	Urine	Feces
Subject A	42. 8	57. 1	34. 3	65. 7	31. 7	68. 3
Subject B	52. 4	47. 6	24. 7	75. 3	X	X
Subject D	23. 4	76. 6	17. 3	82. 7	17. 0	83. 0
Subject E	9. 3	90. 7	7. 8	92. 2	X	X

the dietary calcium was lowest, intermediate at intermediate levels of dietary calcium, and lowest on the highest calcium intake level, whether the men were at bed rest or ambulatory. This is mainly the result of the more prominent changes in the level of calcium in the feces, which reflected dietary calcium changes.

Tabel IV summarizes the comparison of calcium levels in feces and urine for the men who participated in the first three bed rest units.

Trends in Bone Mass Changes

The change of bone mass in the central section of the os calcis generally followed the level of calcium in the diet, although it is understood that this one section of one bone cannot represent what is happening to the entire skeleton. The ranges of bone mass loss as represented by this central section of the os calcis, which is the width of the scanning beam (1.3 mm.), from a posterior to an anterior landmark in the os calcis were the following for the specified bed rest periods which have been completed.

Bed rest unit	Duration of unit	Level of dietary calcium (grams)	Range of bone mass change in os calcis section
	Days		*Percent*
1	14	1. 5	−4.5 to −6.7
2	14	. 7	−7.4 to −9.8
3	14	2. 0	−3.0 to −6.5
4	14	. 5	−10.6 to −14.0
5	30	2. 0	−3.5 to −7.4

It is noteworthy that the group which exhibited the highest level of negative calcium balance (unit 4 on 0.5 gram calcium offered, with a mean of 0.432 gram consumed) also experienced the highest loss of bone mass in the central os calcis section during a 14-day bed rest period. The high negative calcium balance values, which ranged from −0.507 to −0.813, as shown in

table III, are consistent with the high percentages of bone mass loss, as shown above.

Changes in the bone mass of finger-phalanx 5–2, determined by scanning cross sections 1 mm apart throughout the length of this bone, were minor during all 14-day bed rest immobilization periods. During the 30-day bed rest unit which is completed, however, over-all losses in bone mass in this anatomical site ranged from −7.6 to −11.9%. Most of the losses in bone mass in this position occurred during the last half of the 30-day bed rest period.

Changes in bone mass of the patella are being measured at the present time, with no report on the effect of bed rest immobilization on the integrity of this bone completed at the present time.

Changes in Serum Calcium During Bed Rest

Changes in serum calcium have been very slight during all of the bed rest immobilization periods which have been completed to date. Even during the 14-day bed rest period when only 0.5 gram calcium was offered with a mean slightly below this level consumed, serum calcium levels remained relatively constant, as seen in the following summarization:

	Serum calcium (*mg per 100 ml serum*)	
	Initial	*Final*
Subject G	11. 5	11. 3
Subject H	10. 3	10. 3
Subject J	10. 7	10. 6
Subject K	10. 1	10. 7
Subject L	11. 2	10. 5

SUMMARY

Our studies on *calcium loss during human bed rest* involve seven bed rest periods with four to five healthy adult males participating in each bed rest unit. Each bed rest unit involves a preconditioning and a reconditioning ambulatory period and collateral ambulatory periods during which the same level of calcium is fed as during the bed rest phase. As a part of the experimental ambulatory unit, the subjects also were preconditioned and reconditioned.

Results of the bed rest and ambulatory phases of the investigation are reported in terms of dietary balance calculations, changes in bone mass in certain anatomical sites as determined by radiographic bone densitometry, and changes in certain blood values.

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REFERENCES

BIRKHEAD, N. C.; BLIZZARD, J. J.; DALY, J. W.; HAUPT, G. J.; ISSEKUTZ, B., JR.; MYERS, R. N.; and RODAHL, K.: (Monitored by Paul LaChance), Cardiodynamic and Metabolic Effects of Prolonged Bed Rest, Technical Documentary Report No. AMRL–TDR–63–67 (1963) Biomedical Laboratory, Air Force System Command, Wright-Patterson Air Force Base, Ohio, 1963.

CANTAROW, ABRAHAM; and SHEPARTZ, BERNARD: Biochemistry, Third ed., W. B. Saunders Co., Philadelphia, 1962.

DEITRICK, JOHN E.; WHEDON, G. DONALD; and SHORR, EPHRAIM: The Effects of Immobilization Upon Various Metabolic and Physiologic Functions of Normal Man. Am. J. Med., vol. 4, 1948, p. 3.

HOWARD, JOHN EAGER; PARSON, WILLIAM; and BIGHAM, ROY S.: Studies on Patients Convalescing from Fracture: III. The Urinary Excretion of Calcium and Phosphorus. Johns Hopkins Hosp. Bull., vol. 77, 1945, p. 291.

VOGT, F. B.; MACK, P. B.; BEASLEY, W. G.; SPENCER, W. A.; CARDUS, D.; and VALBONNA, C.: The Effect of Bedrest on Various Parameters of Physiological Function: Part XII, The Effect of Bedrest on Bone Mass and Calcium Balance. Prepared under Contract No. NAS 9–1461 by Texas Institute for Rehabilitation and Research, Houston, Texas, for National Aeronautics and Space Administration (April 1965).

WHEDON, G. DONALD: Osteoporois: Atrophy of Disuse, Chap. 4 of Bone as a Tissue, pp. 67–82, McGraw-Hill Book Co., Inc., 1960.

WHEDON, G. DONALD; DEITRICK, JOHN E.; and SHORR, EPHRAIM: Modification of the Effects of Immobilization upon Metabolic and Physiologic Functions of Normal Men by the Use of an Oscillating Bed. Am. J. Med., vol. 6, 1949, p. 684.

WHEDON, G. DONALD; and SHORR, EPHRAIM: Metabolic Studies in Paralytic Acute Anterior Poliomyelitis II. Alterations in Calcium and Phosphorus Metabolism. J. Clin. Invest., vol. 73, 1957, p. 966.

WOHL, MICHAEL G.; and GOODHART, ROBERT S.: Modern Nutrition in Health and Disease. Lea and Febiger, Philadelphia, 1960; based on the report by Tibbetts, Dorothy M., and Aub, Joseph C.: Magnesium Metabolism in Health and Disease: I. The Magnesium and Calcium Excretion of Normal Individuals. J. Clin. Invest., vol. 16, 1937, p. 491.

Regression Curves for Representative Urinary Calcium and Bone Mass Values

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Regression curves are presented as calculated and plotted by the IBM 1620 Computer from urinary calcium excretion and bone mass data obtained from men in the TWU bed rest and collateral ambulation studies. Dr. Klapper has based her Ph. D. research problem on the units of this overall investigation, which included the feeding of 0.7, 1.5, and 2.0 grams of calcium, respectively, during three bed rest and three corresponding ambulatory periods to four healthy adult male subjects shown in the previous report as belonging to Bed Rest Units 1, 2, and 3. Three figures have been chosen to illustrate how the regression curve can serve to show trends in bone mass change and change in urinary excretion of calcium.

FIRST ORDER REGRESSION PLOTS

First, regression lines have been calculated and plotted by the computer based on a program which includes the first order computation of least squares, following the formula:

$$Y = A_0 + A_1(x)$$

where

Y is the dependent variable (urinary calcium or bone mass);

x represents time;

A_0 is the Y intercept; and

A_1 is the slope of the regression line.

Simultaneously with plotting the regression lines, the data points also were plotted by the computer. The following data were obtained from the machine in each case, as follows: (a) the *variables* involved; (b) the *range* covered by the plot; (c) the *constant*, which is the same as the intercept in analytical geometry; (d) the *slope* of the curve, which denotes the rate of change of the factor being plotted; and (e) the *standard error of estimate.*

These data have enabled the plots to be drawn and interpreted. In each linear plot that has been produced, the standard error of estimate has indicated that the line which was plotted is a reasonable approximation of the change which has occurred with time in the factor involved.

COMPARISON OF LINEAR BONE MASS REGRESSION PLOTS FOR A REPRESENTATIVE SUBJECT INVOLVING THREE BED REST PERIODS

Data from Subject A were chosen to show in figure 1 the slopes of the computer curves during three bed rest periods when three different levels of calcium were being fed.

Upper Plot.—The first order regression line is plotted against time for the bone mass measurements in the central section of the os calcis obtained while Subject A was experiencing 14 days of bed rest (Bed Rest Period 2) with the lowest level of dietary calcium consumed (0.7 gram per day planned and a mean of 0.659 gram consumed). The data points for the 14 days of the experiment also are shown in this and subsequent figures.

Middle Plot.—The same data are shown for Subject A, measured during Bed Rest Period 1 (1.5 grams dietary calcium planned and a mean of 1.457 grams consumed).

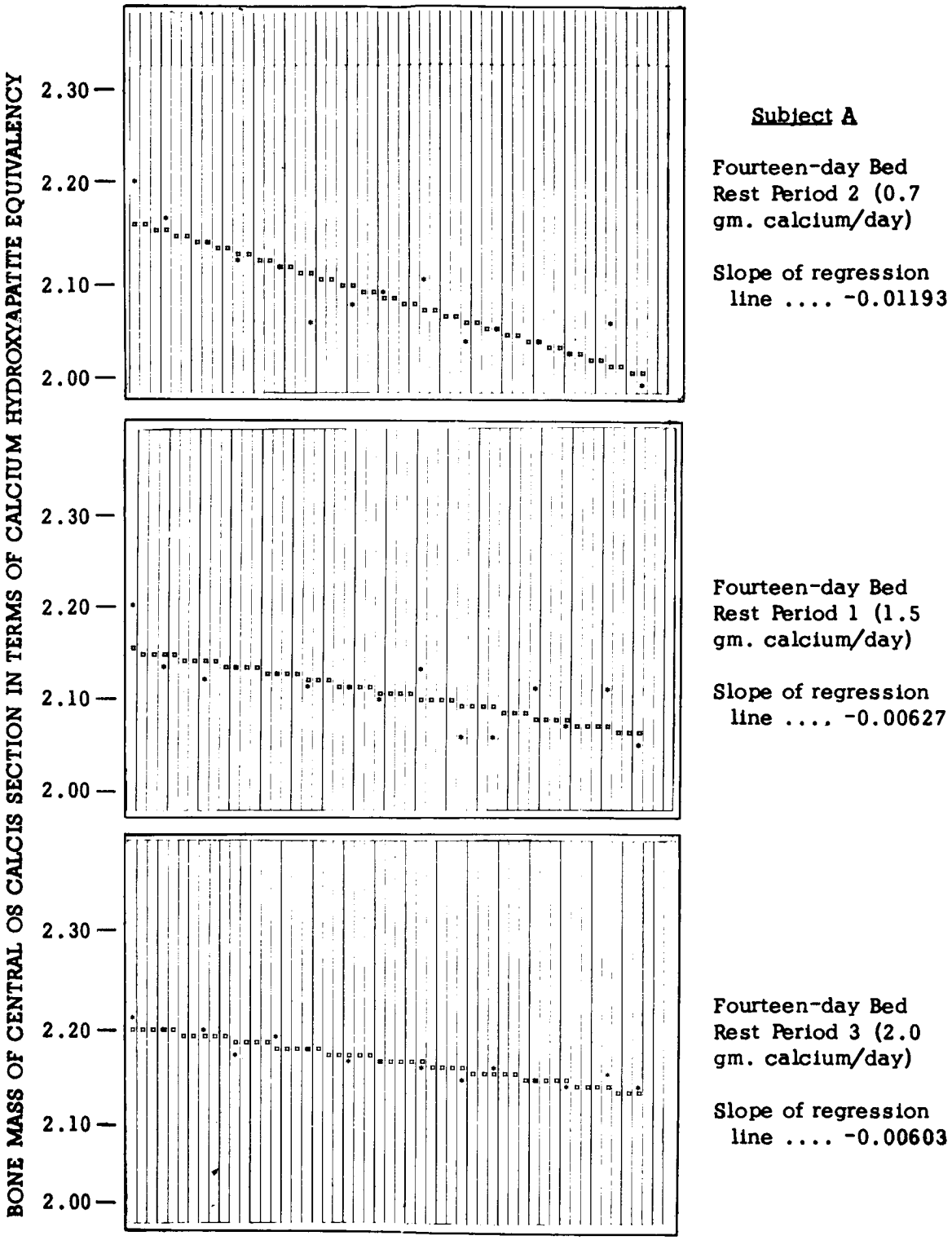

FIGURE 1.—Comparison of linear bone mass regression plots for a representative subject involving three bed rest periods—Subject A.

Lower Plot.—The same data are graphed for Subject A during Bed Rest Period 3 (2.0 grams dietary calcium planned and 2.017 grams consumed).

The slopes of the three regression lines for bone mass of this subject in the anatomical site specified are shown in each of the subdivisions of figure 1.

In order to test the hypothesis that the regression coefficient (A_1 in the formula on p. 179) which was obtained during a bed rest or an ambulatory period was equal to some specific value, a "t" test was run to find whether or not this differed significantly from zero. It was found that, for all 14-day bed rest periods for Subject A, the negative slopes of the regression lines were less than zero by differences which were highly significant in all three cases. The same type of significant differences was found for each bed rest period for the remaining subjects in the three units of the investigation under discussion.

COMPARISON OF FIRST ORDER BONE MASS REGRESSION PLOTS FOR A SUBJECT DURING BED REST AND DURING AMBULATION ON A DIET PROVIDING 0.7 GRAM CALCIUM DAILY

Subject B was used as an example to illustrate the comparison of first order bone mass regression lines during 14 days of bed rest and during the same length of time when he was ambulatory, while a diet was consumed which provided 0.7 gram of calcium daily. When this subject was at bed rest with 0.7 gram of calcium provided daily, he consumed a mean of 0.636 gram. When he was ambulatory on the same diet, he averaged 0.649 gram daily. Therefore his calcium consumption was virtually the same during the two periods.

The computer gave the slope of the regression line in the upper plot of figure 2 as -0.00898, while the middle plot showed the slope of the line to be -0.00233. The application of the "t" test showed that the regression line both for the subject's bone mass during bed rest and during ambulation had specific values which differed from zero by statistically significant differences. Significant "t" values were found for the other three subjects of the group under discussion not only for the bed rest but also for the ambulatory period when 0.7 gram of calcium per day was fed.

It was not expected that a negative calcium balance would be found for subjects while ambulatory on 0.7 gram of calcium per day. The idea that excessive sweating may have resulted in calcium losses which were not measured was abandoned because the time of year when this unit was accomplished (December) was not conducive to the loss of much perspiration. Since other parts of the study had indicated that, when the feeding of a low dietary calcium level followed immediately after a higher level, the tendency for bone mass to decrease even when the subject was ambulatory may have some bearing here, since this group was equilibrated on a diet containing 1.5 grams of calcium immediately before this ambulatory period began.

REPRESENTATIVE BONE MASS REGRESSION PLOT FOR A SUBJECT AMBULATORY WHILE IN 1.5 GRAMS CALCIUM PER DAY

For comparative purposes, a computer plot for Subject B while he was ambulatory on the equilibration diet which provided 1.5 grams of calcium daily is shown in the bottom plot of figure 2. The slope of this regression line was $+0.00013$, and the application of the "t" test showed that the line did not differ significantly from zero. These findings characterized the linear regression plots for all four subjects of this group during an ambulatory period when 1.5 or 2.0 grams of calcium were provided.

EXPONENTIAL REGRESSION PLOTS

As a means of studying the type of exponential regression curves which could be obtained by means of the IBM 1620 Computer, semilog curves were plotted for urinary calcium excretion and bone mass change values, with data which seemed to define the results in a manner which supplemented the linear regression plots.

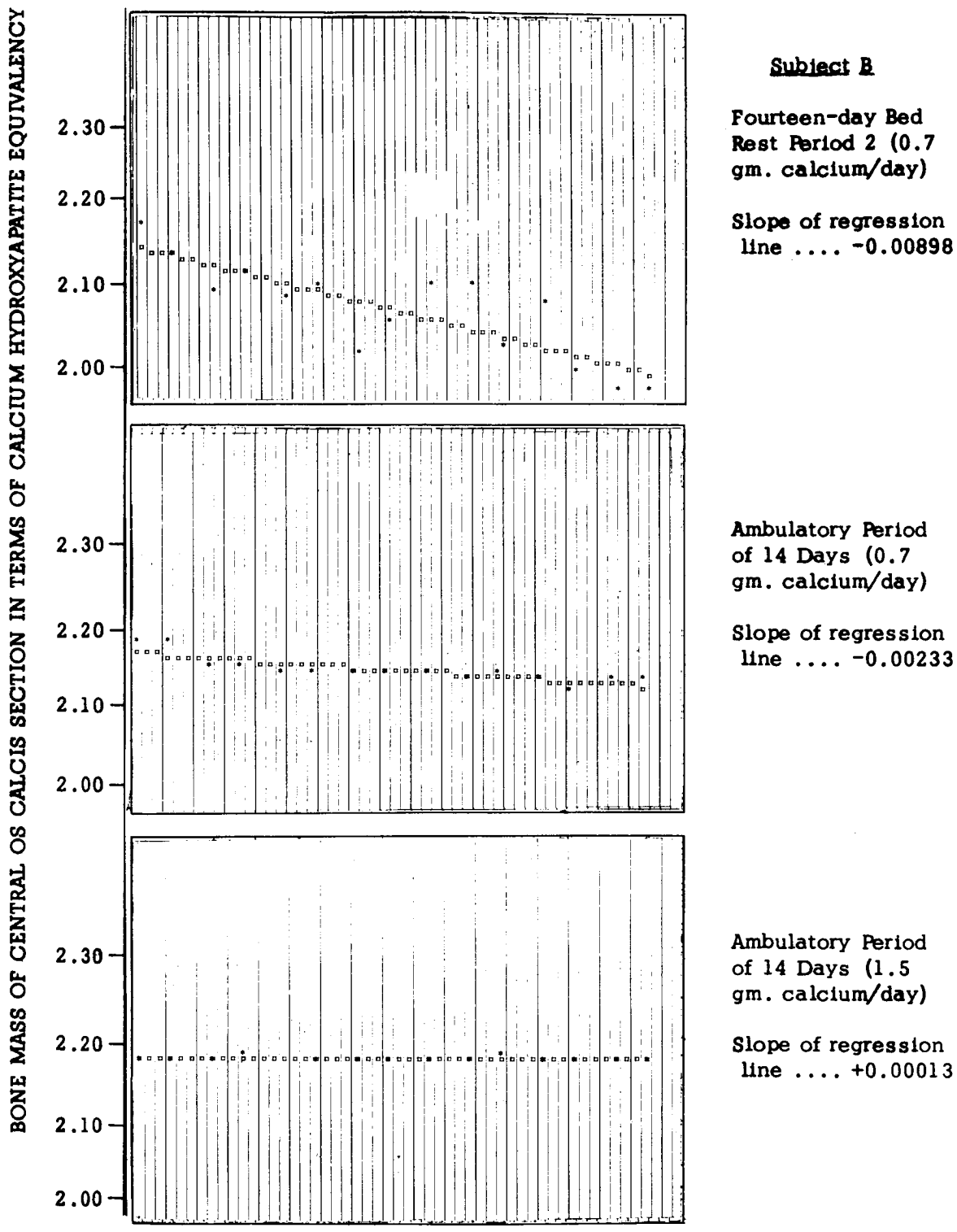

FIGURE 2.—Comparison of linear bone mass regression plots for a representative subject involving three bed rest periods—Subject B.

The semilog curves were based on the following formula:

$$Y=A_0+A_1 \log (x+1)$$

where

Y is the dependent variable;
x represents time;
A_0 is the intercept;
A_1 is the exponential factor.

Again, data points are computed and plotted by the computer at the same time that the semilog curves are made. The semilog curves are plotted in such a way that the dependent variable data points have the same scale as in the linear plots of the same data. Since the dependent variable ordinate has a linear scale, the least squares semilog curve appears as a log curve. The standard error of estimate, however, is not the normally defined standard error. Rather, it is the antilog of the standard error of log $(x+1)$.

COMPARISON OF A LINEAR COMPUTER PLOT OF CALCIUM URINARY EXCRETION WITH EXPONENTIAL PLOTS OF URINARY CALCIUM EXCRETION AND BONE MASS CHANGE

Figure 3 is presented to show a comparison of a linear with an exponential plot of the same variable (urinary calcium excretion), and of the exponential plots of two variables (urinary calcium excretion and bone mass change).

The figure is based on the urinary calcium and the bone mass data for Subject D of this group of subjects when he was on bed rest with 0.7 gram of calcium provided daily and a mean of 0.686 gram consumed.

The slope of the regression line for the urinary calcium loss was +0.13406 units and the probability that this slope differed significantly from zero was <0.025. The urinary calcium values are plotted on a daily basis, and are not averaged for a group of succeeding days as is the custom of many investigators. Therefore, the values show a wider range than if the means for a period of several days were plotted.

The trends shown by the linear and the exponential computer plots for urinary calcium were similar for all four men of this group for this bed rest unit, as was the exponential plot for the bone mass values during the same bed rest period. The figures shown here are only intended to be representative.

One section of one bone involved in these computer plots by no means represents the changes which are taking place in the entire skeleton. Nevertheless, the trend of negative change shown in the bone mass plots bears a relationship to the trend of excretion of calcium in the urine, even though the total weight of this element lost in this bone section is not presumed to be equal to that lost in the urine.

A comprehensive bulletin is being prepared for publication by the Texas Woman's University presenting data and graphs for all subjects for all parts of all units included.

ACKNOWLEDGMENTS

The authors wish to express their gratitude to Mr. John Hornbuckle, Director of the TWU Data Processing Center, for assisting with the statistical analysis of the data involved in this brief preliminary report. Thanks also are due to Jessie Ashby, Reba Fry, and Lenoir O'Rear, for coding and otherwise preparing the data for computer processing.

COMMENTS

Dr. NORDIN. In what order were these balances done?

Dr. KLAPPER. These are different subjects.

Dr. NORDIN. The previous one was the same subject?

Dr. KLAPPER. Yes, the 1.5 gm intake was done first. The 0.7 was done second and third purposely.

Dr. NORDIN. With regard to figure 3, in the lower two graphs, which is the log and which is the semi?

Dr. MACK. That is not a log scale. In the upper two graphs the dots are the actual values for the data points for the urinary calcium.

Dr. NORDIN. Urinary calcium does not change by 100 mg a day from one day to the next. There must have been some urinary collection error or, possibly, some analysis error.

Dr. KLAPPER. This goes up to 400 mg a day. This is the actual expression in hundreds of milligrams.

Dr. NORDIN. It is not a semilog plot. It is arithmetic. Which is the semilog, Dr. Klapper?

Dr. MACK. The semilog was done by the computer when it made the regression line.

Dr. RICH. I gather that the squares are the com-

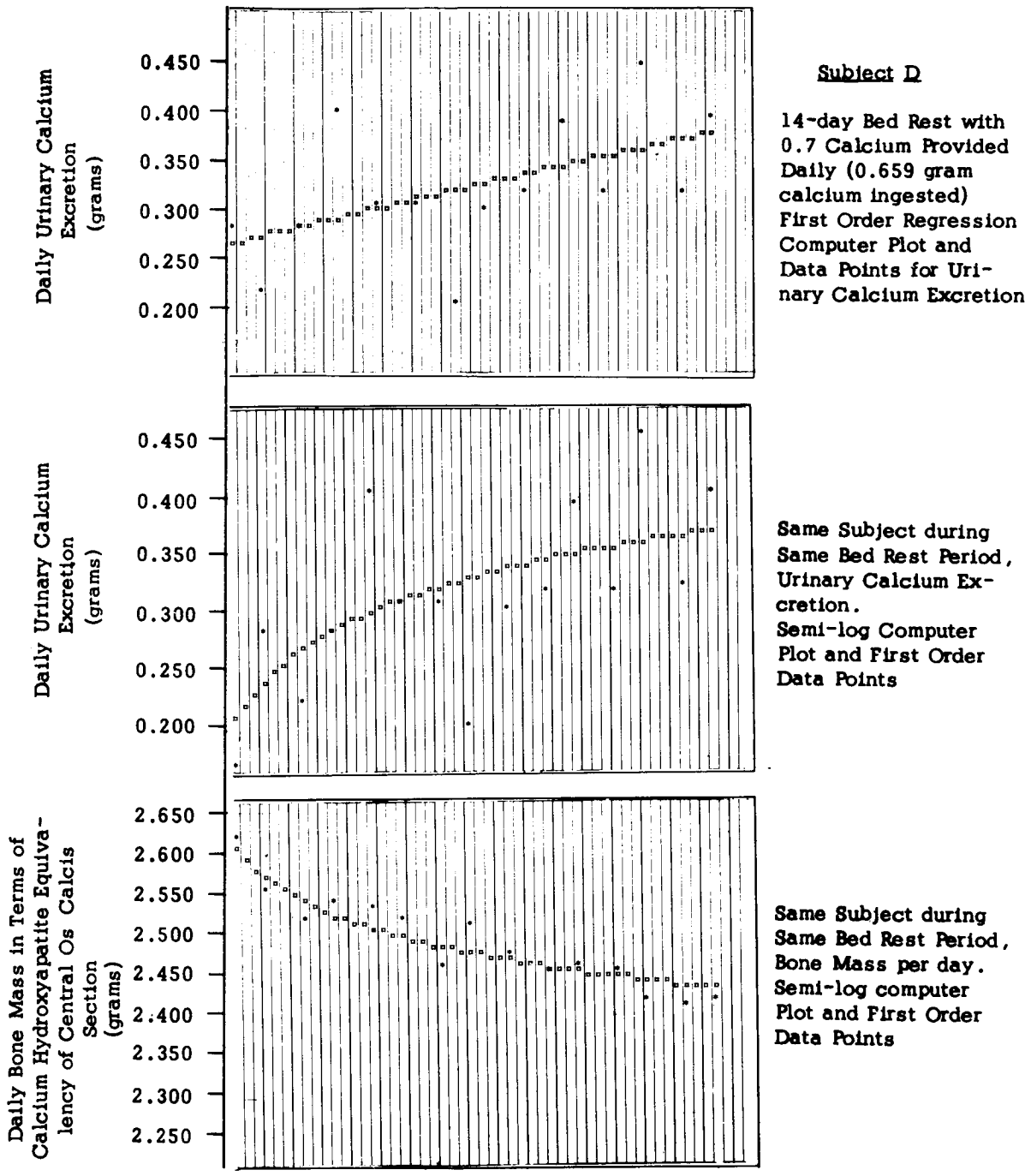

FIGURE 3.—Comparison of a linear computer plot of calcium urinary excretion with exponential plots of urinary calcium excretion and bone mass change.

puter's running average from the individual data? Are these the black dots that are hard to see?

Dr. KLAPPER. Yes.

Dr. MACK. The black dots are the actual data points. That is the computer semilog curve.

Dr. RICH. The earlier slide of the very early change in the density of the os calcis, those showing very uniform straight lines, I presume also those are the computer average of the actual figures.

Dr. MACK. They are the computer logs based on the equation of least squares.

Dr. ROCKOFF. How many films analyzed are represented by each dot?

Dr. MACK. One film taken once but analyzed four times for each day. There was a film a day unless one was skipped accidentally for some reason. When we were preparing for this, which we did for several months, we took two and three films a day, and it required too much radiation on the patient. One film a day analyzed four times has been our standard. We deviate from that for certain research purposes.

Dr. UEIST. Dr. Mack, you have a very important clinical observation here. We want to compare your results with those of Dr. Whedon and with John Eager Howard's patients who were immobilized. At the bottom of this curve that showed the negative calcium balances, what was the magnitude of the negative calcium balance at the end of 14 days in these patients?

Dr. MACK. The greatest one was —0.813 gram mean balance for the 14 days.

Dr. UEIST. I wonder how this compared with Dr. Whedon's values on paralyzed polio patients and John Howard's values on patients immobilized in treatment of fracture.

Dr. WHEDON. I think a more pertinent point would be not just knowing about the greatest change, but what the change was in the various subjects and some idea of the mean change (mean daily negative balance for first 14 days: 597 mg for five subjects at 500 mg intake; 313, for four at 700; 174, for four at 1500). Nevertheless, to indicate an order of magnitude, the shift in calcium balance during about 14 days in the four normal subjects, immobilized in plaster (Deitrick et al.; 1948), would be of the order of 100 to 200 mg per day (135 mg/day, range 98 to 168). As you will recall, our subjects were kept at bed rest for 6 and 7 weeks and the increase in calcium loss, the negative of calcium balance steadily increased until the fifth or sixth week (mean daily negative balance for 6 to 7 weeks immobilization: 236 mg, range 156 to 380).

The level of shift at 2 weeks is only part way along the curve. At the same level of intake, 920 mg, 5 paralytic polio patients had a mean daily negative calcium balance during the second to third months of 439 mg, range 277 to 575.

Dr. MACK. Dr. Whedon, that value I gave was for my subjects on 0.5 of a gram intake. You had none that low?

Dr. WHEDON. This is a very good point. Dr. Mack has been doing a series of studies at different calcium intake levels. Our studies of normal subjects were all at the same calcium intake levels which was 920 mg of calcium per day.

Dr. SMITH. Do I understand that you were giving 10,000 units of vitamin D? That is approaching a level where there may be a primary action of vitamin D on the bone.

Dr. MACK. That is vitamin A equivalency and takes in carotene. Vitamin D was 400 units.

Dr. RICH. The very interesting aspect of your study is the correlation of the actual bone mass changes with the balance studies reported previously. It is intriguing that you find that your patients who lose the most calcium lose about 1% of the skeleton per day, if I understand correctly that your coefficients refer to loss from the os calcis. This should cause a loss of about a gram per day from the body in terms of balance results. You are very close if you have a 600 mg loss; it is surprising that it would be that close in two such different techniques. [See postconference note below—ED.]

Dr. WHEDON. I admire Clayton Rich's desire to bring about a correlation here, but I think it is quite risky. Remember, this is just the os calcis and the rate of loss from trabecular bones could be exceedingly difficult to assess and should be greater than for the total skeleton.

Dr. RICH. I agree. I was pointing out the similarity of figures. It is an order of magnitude.

[*Editor's postconference note:* This attempted correlation is of great interest. In fact, however, the os calcis density loss reported by Dr. Mack was 10–14% in 2 weeks for the 0.5 g calcium intake group, rather than 1% as Dr. Rich thought. The densitometric rate of loss, when applied to the whole skeleton containing 1000 to 1500 g of calcium, would predict a loss of 100 to 200 g total for the 2-week period, or 7 to 14 g mean loss per day. Since, in contrast, the mean loss for this group by balance study was 0.5 to 0.8 g per day, the correlation is not close and would tend to support the idea that the considerably trabecular os calcis loses calcium at a much greater rate (10 to 15 times) than the average for the whole skeleton. The same general ratio appeared to hold at the 2.0-g intake level where the os calcis density loss was 3 to 6% and the mean daily calcium balance —0.2 to —0.32 g. Examination of this association in on-going and future studies is potentially of considerable value.—D.W.]

Dr. MACK. Again, that loss of nearly 1 g a day is from the lowest level of calcium that we feed.

Dr. Whedon, I did want to mention that you had some slides at a previous conference in which you did show a correlation between calcium intake level and urinary loss.

Dr. WHEDON. Dr. Mack is referring to studies that we have done with patients with osteoporosis in which we could influence the calcium balance significantly by varying the calcium intake.

Normal "Osteoporotic" Bone Loss

STANLEY M. GARN, CHRISTABEL G. ROHMANN, ELEANOR M. PAO, AND ETHEL I. HULL
Fels Research Institute

The behavior of cortical bone and peripheral bone during the age-associated period of central bone loss has been the subject of some dissension. There is a common belief that cortical bone does not become reduced in osteoporotic bone disease, and that peripheral bone is spared until very late in the disease. Some of these statements have been made in reference to experimental studies on rats (for example, the work of Ellinger et al., 1952, as quoted by Malm, 1958), or in reference to the sheep experiments of Benzie et al., 1956. Customary clinical concentration on the lower spine undoubtedly contributes to this opinion (cf. Gershon-Cohen et al., 1953).

On the other hand, there is growing evidence that cortical bone, peripheral bone, and even nonweight-bearing bone participate in the process of bone loss. Such evidence comes from dry, defatted skeletons and ashed skeletons (see Trotter, 1954; Baker, 1964), from radiographic studies of the arm, elbow, and femur (Meema, 1963; Meema and Meema, 1963; Smith, 1964; Smith et al., 1964 a, b; Barnett and Nordin, 1960; Nordin, 1965). These well documented studies indicate that peripheral cortical bone loses with age and in osteoporosis.

In view of the technical difficulties in *in vivo* clinical or field appraisal of bone loss in the spine, it is of particular interest to ascertain the extent to which bone loss takes place in the hand, which is readily accessible to radiography and quite suitable for a variety of techniques of bone measurement. The present paper is concerned with the measurement of bone loss in the hand, in individuals of three racial groups, and in serial, longitudinal, as well as in cross-sectional, context.

METHODS AND MATERIALS

The present study is based upon cross-sectional and longitudinal measurements of cortical thickness on the second metacarpal, in a total of nearly 1,000 subjects, 25 to 101 years of age. The subjects, unselected with respect to bone disease, included over 600 volunteer participants in long-term studies in growth and development in the Fels Research Institute, including members of the Senior Citizens Group of Yellow Springs, 60 subjects of Japanese and Chinese ancestry resident in southwestern Ohio who participated in studies of the relationship between diet and bone loss, and over 200 skeletalized Negro subjects of known age, sex, and cause of death, from the Terry collection of Washington University School of Medicine, St. Louis, Mo.

Measurements of cortical thickness were made with pinpoint calipers on standardized postero-anterior radiographs of the left hand, reading out to the nearest 0.1 mm or better (cf. Garn et al., 1963, 1964). Intraobserver and interobserver replicability of the measured values exceed 0.98, as previously described in this Symposium. Error analysis of interobserver differences in the longitudinal part of the study showed that the RMS error attributable to interobserver difference was approximately 0.15 mm.

In addition to the cross-sectional 4-decade comparison of cortical thicknesses, it was also possible in over 49 women and 26 men to make

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TABLE IA.—*Age-Associated Loss—Cortical Thickness of the Second Metacarpal*

Age	Ohio whites						Missouri Negro skeletons					
	267 Males			352 Females			111 Males			117 Females		
	N	$\bar{X}$	SD	N	$\bar{X}$	SD	N	$\bar{X}$	SD	N	$\bar{X}$	SD
25–34	62	5. 9	0. 6	153	5. 4	0. 8	24	5. 3	0. 7	24	4. 8	0. 7
35–44	92	5. 8	0. 7	85	5. 5	0. 7	24	5. 2	0. 6	25	4. 6	0. 5
45–54	60	5. 7	0. 7	61	5. 2	0. 7	22	4. 6	0. 6	22	4. 0	0. 6
55–64	34	5. 3	0. 6	31	4. 6	0. 6	20	4. 7	0. 6	20	3. 9	0. 8
65–[a]	19	4. 7	0. 4	22	3. 7	0. 7	21	4. 3	1. 0	26	3. 3	0. 7
Decrease	20%			31%			19%			31%		

[a] Mean age: males 72, females 74—March 1965 data.

TABLE IB.—*Age-Associated Loss—Cortical Area of the Second Metacarpal*

Age	Ohio whites				Missouri Negro skeletons			
	267 Males		352 Females		111 Males		117 Females	
	N	$\bar{X}$	N	$\bar{X}$	N	$\bar{X}$	N	$\bar{X}$
25–34	62	59. 6	153	44. 7	24	57. 1	24	43. 6
35–44	92	58. 5	85	45. 7	24	53. 3	25	45. 1
45–54	60	59. 7	61	43. 8	22	50. 7	22	39. 0
55–64	34	58. 5	31	42. 2	20	53. 1	20	38. 5
65	19	50. 6	22	36. 6	21	48. 8	26	34. 1
Decrease	15%		18%		15%		22%	

TABLE IC.—*Age-Associated Loss—Cortical Ratio of the Second Metacarpal**

Age	Ohio whites				Missouri Negro skeletons			
	267 Males		352 Females		111 Males		117 Females	
	N	$\bar{X}$	N	$\bar{X}$	N	$\bar{X}$	N	$\bar{X}$
25–34	62	63	153	67	24	56	24	59
35–44	92	62	85	68	24	56	25	54
45–54	60	60	61	66	22	49	22	49
55–64	34	54	31	57	20	50	20	47
65	19	50	22	46	21	46	26	39

* Following Nordin ratio cortex/total periosteal diameter. Note cortical ratio is initially higher in females.

long term individual comparisons of bone loss, generally between the 4th and 5th decade and the 6th, 7th, and 8th decades. Individual spans between successive X-rays were not less than 15 years, and in some cases included a time lapse of 30 years or more.

FINDINGS

As shown in table I, cortical thickness decreased after the 5th decade in both sexes and in Negroes and whites. For Ohio white males and for Missouri Negro males the amount of bone loss approximated 20%. For Ohio white females and Missouri Negro females alike, the amount of bone loss approximated 31%. Analysis of Japanese and Chinese data indicated a comparable age-associated loss of bone ($r = -0.41$), greater for the females than for the males. Though the race differences in the amount of compact bone were considerable, with the Chinese and Japanese subjects evidencing far less compact bone as previously shown at all ages (Garn, et al., 1963), there was no evidence of differences among these three populations in rates of bone loss with age.

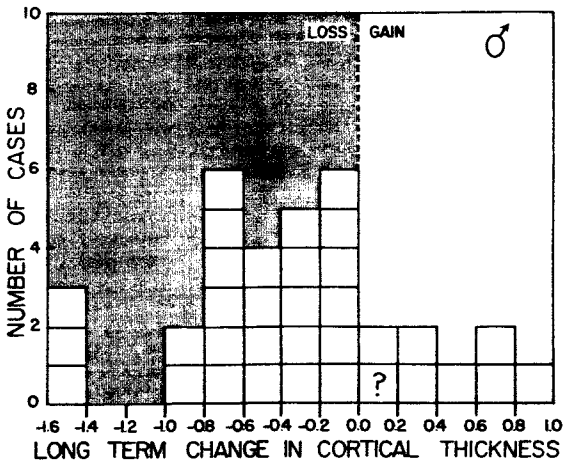

FIGURE 1.—Long term cortical bone loss in 42 women and 34 men. Over a span of 15–32 years, the majority of subjects of both sexes lose considerable compact bone. Only a few women and a limited number of men gained bone in excess of the RMS measuring error of 0.15 mm. While it is clear that the majority of individuals lose bone between the 4th and the 7th decades, there are systematic differences that set off the losers from the gainers.

Longitudinal analysis of individual male and female participants in the Fels studies in growth and development further confirmed both the universality of bone loss with age and the age at onset. The vast majority of subjects of both sexes lost bone between the 4th and 5th, and 6th and 7th decades as shown in figure 1. However, as indicated for the 49 females followed in complete longitudinal context, and as previously observed for males from the same subject population, there are some individuals who do not appear to lose bone with age; and, in fact, they actually gain bone. In addition to the individuals shown, some subjects (such as F–1) actually gained bone over 3 successive decades. On the other hand, the majority do lose bone; and in extreme cases, the loss of compact bone in the hand is so great as to be perfectly apparent in radiographs taken only a few years apart (see figs. 2 and 3).

In summation, cortical bone of the hand is lost in American white, American Negro, and Oriental subjects of both sexes, to the same extent, far more in females than in males; and this generalization holds for the majority of individuals within the population as well as for the population itself. Studied longitudinally, bone loss is not a phenomenon of a few osteoporotic extremes, but rather a characteristic of most individuals in all groups.

DISCUSSION

It is now abundantly clear that cortical bone in the hand is lost progressively after the 5th decade, in both sexes and in whites, Negroes, and Orientals at about the same rate. With no decrease in the periosteal diameter[1] and, therefore, the periosteal volume, the cortical thickness of the second metacarpal and the absolute bone volume decreases 20% on the average in males. The rate of loss over the same time period is greater (30%) in females.

[1] Periosteal diameter remained essentially constant (7.83 vs 7.94 mm). However, by sign test, 35 out of 49 women gained in periosteal diameter from the earlier to the later ages sampled. Clearly, the reduction in cortical thickness was not due to reduced periosteal diameter.

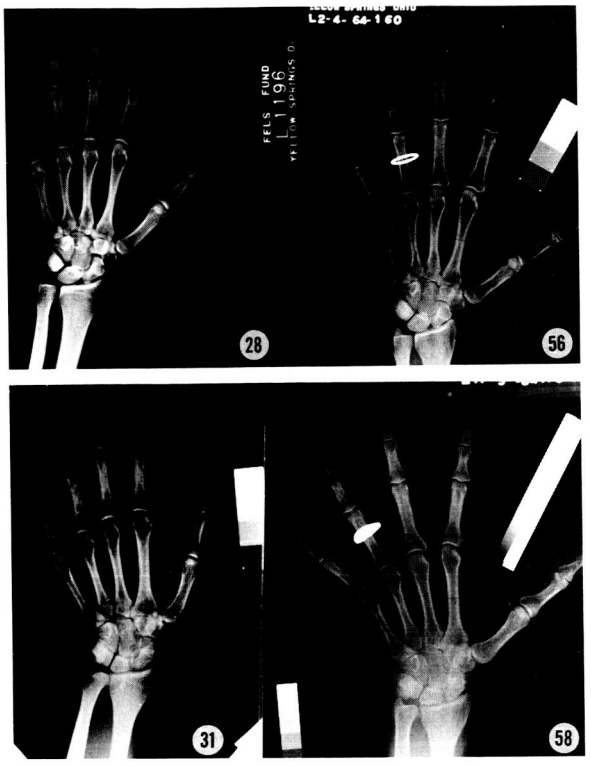

FIGURE 2.—*Above:* 1.6 mm of cortical loss in 28 years. Although the 3-decade bone loss here is 24%, this woman (MO 160) started with a well above average amount of cortical bone at age 28 (6.9 mm) and at 56 years of age still has an above average amount of cortical bone (5.3 mm). She is thus protected against mechanical bone failure for another 2 decades. *Below:* Bone loss in subject MO 49 over a 27-year period. Starting with 5.3 mm of cortex at age 31, she lost 1.2 mm (23%) by age 58, at which age she experienced numerous fractures. In general, women who have below-average amounts of corical bone to start with, and who lose average or above average amounts of cortical bone are then more subject to mechanical bone failure.

The female, with considerably less bone to begin with, also loses bone faster to the point where mechanical integrity may be impaired. Though we had previously believed that "osteoporotic" bone loss was less common in Negroes, and possibly rarer in Orientals, the virtual identity of the percentage losses in the close to 1000 individuals represented in this study attests to interpopulation universality of compact bone loss.

Individual rates of bone loss have been studied over a long time period, in some cases as long as 35 years. Comparing individuals in their 3d and 4th decades to the same individuals in their 6th and 7th decades, the majority of subjects of both sexes lose bone. For the purposes of defining osteoporosis, however, the extremes of individually determined bone loss must be distinguished. In static or cross-sectional studies, there is inevitably confusion between those individuals who have little bone (i.e., a thin cortical wall) and those individuals whose cortical deficiencies are due to bone loss. In longitudinal perspective, it is possible to separate the nonlosers (or even bone gainers) after the 5th decade from those who have lost 20 to 30% of compact bone in the period of 15 years or so. It is the latter who constitute true osteoporotics, who then pose particular problems for metabolic study.

The loss rates for cortical bone in the hand found in the present study agree with other findings on the hand (Smith, 1964; Smith and Walker, 1964) and on the elbow (Meema, 1963; Meema and Meema, 1963; Nordin, 1965; and others). They parallel—in order of magnitude—loss rates for other tubular bones, and they are completely in line with measurements of bony loss in supporting bone made densitometrically on the first metatarsal (Garn et al., 1965b).

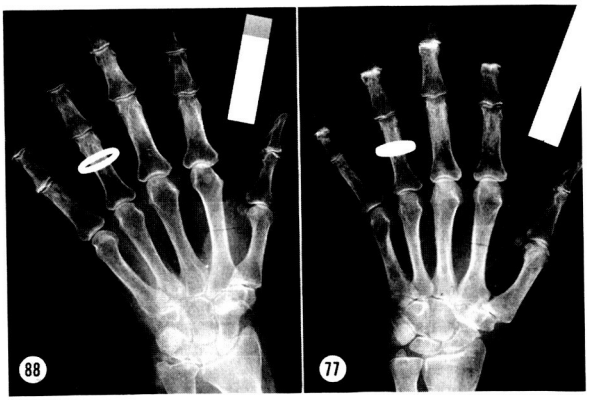

FIGURE 3.—Radiographs of two women: one 88 years of age (left), and the other 77 years of age (right), showing extreme loss of cortical bone. In both of these women, loss of cortical bone has proceeded to the point where the entire thickness of the cortex is below expectancy for a 4-year-old girl from the same subject population. For comparative data see Garn et al. (1963) and Garn et al. (1965).

Cortical bone does participate in the age-associated process of bone loss, and peripheral as well as central bone is lost. Weight-supporting bone appears to be lost as rapidly as the non-weight-bearing bone of the upper extremity. After reviewing many aged hands in which cortical thickness of the second metacarpal is reduced to infant values, in which partial collapse of cortical bone can be seen, and in which the metacarpals themselves have lost space, and after viewing the feet of the same subjects, it is impossible to agree that "osteoporosis" is primarily a disease of the central skeleton. We cannot agree that bone loss, though greater, is not restricted to the female.

Extrapolating the values we have observed to the entire skeleton, "normal" or, more specifically, average rates of bone loss appear to be between 5 and 10% per decade. Under these circumstances, the average subject in this age span must be losing about 15 to 30 mg of calcium per day. The loss rate is probably proportional to the skeletal mass itself, since the rate of bone loss is proportional to the initial cortical thickness. However, loss of 15 to 50 mg of calcium a day would obviously be more serious for a small female with a 2- to 2.5-kilogram skeletal mass than to a large and heavy-boned male with a total skeletal mass possibly as great as 5 kilograms.

SUMMARY

The age-associated loss of cortical bone at mid-shaft on the second metacarpal was investigated in over 600 living Ohio whites, over 200 American Negro skeletons from the Terry Collection at Washington University, and in 60 living individuals of Chinese and Japanese descent.

Despite population differences in the average thickness of cortical bone, the loss in thickness approximated 20% in males and 30% in females of each group, with the bulk of the loss taking place after the 5th decade. Expressed as cortical area (corresponding more nearly to the Absolute Bone Volume of Frost), loss rates were proportional.

Explored on a purely *longitudinal* basis over a 15- to 30-year period, it was clear that bone loss was characteristic of the vast majority of aging individuals of both sexes. Indiviual long-term loss rates pointed to a very few who did not lose bone and a few who lost bone excessively. The latter may be considered as the "true" osteoporotics, distinguishable in longitudinal study from those who have little bone to begin with.

Extrapolated to the entire skeleton, the "normal" loss of bone appears to be between 5 and 10% per decade, indicating, for a reference man or women in the 45 to 65 age group, an average calcium loss of 15 to 30 mg per day.

ACKNOWLEDGMENTS

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REFERENCES

BAKER, P. T.; ANGEL, J. L.; LITTLE, M. A.; and MAZEES, R. B.: Studies in Bone Density: Age, Sex, Race, Altitude and Nutritional Factors. Department of Sociology and Anthropology, The Pennsylvania State University, University Park, Pa. Privately printed, 1964.

BARNETT, E.; and NORDIN, B. E. C.: The Radiological Diagnosis of Osteoporosis. Clin. Radiol., vol. 11, 1960, pp. 166–174.

BENZIE, D.; BOYNE, A. W.; DALGARNO, A. C.; DUCKWORTH, J.; HILL, R.; and WALKER, D. M.: Studies of the Skeleton of the Sheep: II. The Relationship between Calcium Intake and Resorption and Repair of the Skeleton in Pregnancy and Lactation. J. Agric. Sci., vol. 48, 1956, pp. 175–187.

ELLINGER, G. M.; DUCKWORTH, J.; and DALGARNO, A. C.: Skeletal Changes During Pregnancy and Lactation in the Rat: Effect of Different Levels of Dietary Calcium. Brit. J. Nutr., vol. 6, 1952, pp. 235–253.

GARN, S. M.; ROHMANN, C. G.; and NOLAN, P., Jr.: Studies on the Development of Compact Bone in Normal Individuals and in Endocrine and Nutritional Abnormalities. Department of Growth and Genetics, Fels Research Institute, Yellow Springs, Ohio. Privately printed, 1963.

GARN, S. M.; ROHMANN, C. G.; and GUZMAN, M. A.: Malnutrition and Skeletal Development in the Pre-School Child. In: Prevention of Malnutrition in the Pre-School Child, W. Henry Sebrell, ed. Food and Nutrition Board, National Academy of Sciences—National Research Council, Washington, D.C., 1965.

GARN, S. M.; ROHMANN, C. G.; and NOLAN, P., Jr.: The Developmental Nature of Bone Changes during Aging. In: Relations of Development and Aging, James E. Birren, ed. Charles C. Thomas, Springfield, Ill., 1964.

GARN, S. M.; WAGNER, B.; and COLBERT, C.: Unpublished data. Department of Growth and Genetics, Fels Research Institute, Yellow Springs, Ohio, 1965.

GERSHON-COHEN, J.; RECHTMAN, A. M.; SCHRAER, H.; and BLUMBERG, N.: Asymptomatic Fractures in Osteoporotic Spines of the Aged. J. Am. Med. Assoc., vol. 153, 1953, pp. 625–627.

MALM, O. J.: Calcium Requirement and Adaptation in Adult Men. Oslo University Press, Oslo, 1958.

MEEMA, H. E.: Cortical Bone Atrophy and Osteoporosis as a Manifestation of Aging. Am. J. Roentgenol., vol. 89, 1963, pp. 1287–1288.

MEEMA, H. E.; and MEEMA, S.: Measurable Roentgenologic Changes in Some Peripheral Bones in Senile Osteoporosis, J. Am. Geriat. Soc., vol. 11, 1963, pp. 1170–1182.

NORDIN, B. E. C.: The Relation between Dietary Calcium and Osteoporosis in Different Parts of the World. A Report to the Nutrition Section of the World Health Organization. Privately printed, 1965.

SMITH, R. W., Jr.: Osteoporotic Changes in Bone Mass With Age. Presented at the 92nd Annual Meeting of the American Public Health Assoc., Inc., New York, 1964.

SMITH, R. W., Jr.; and FRAME, B.: Concurrent Axial and Appendicular Osteoporosis: Its Relationship to Calcium Consumption. New Eng. J. Med. (in press).

SMITH, R. W., Jr.; and WALKER, R. R.: Femoral Expansion in Aging Women: Implications for Osteoporosis and Fractures. Science, vol. 145, 1964, pp. 156–157.

TROTTER, M.: A Preliminary Study of Estimation of Weight of the Skeleton. Am. J. Phys. Anthrop., vol. 12, 1954, pp. 537–552.

COMMENTS

Dr. ROCKOFF. Is it possible that marked changes in technique over the years could alter the cortical thickness measurement?

Dr. GARN. Do you mean marked technique in FPSKV density? We have examined the effects of density variations per se. Down to the point where they are too light to measure, they do not affect what you are talking about.

Dr. ROCKOFF. How about the kilovoltage on the two studies? This looks like a higher kilovolt study. Does that include kilovolt?

Dr. GARN. Kilovolt changes make a difference in the density equivalent values since they change the ratio of apparent densities between bone and the various type of wedges. They do not in our findings affect the cortical thickness measurement except possibly at very high kV's, which we have not yet evaluated. Our techniques for the hand have been on a screen-type film between 30 and 40 PKV, that is, 30 plus twice part thickness. The nonscreen film technique we are using in connection with densitometry is fixed at 50 PKV at the requirement of the film manufacturer. This variation should not be a problem. However, Dr. Paulus has recently sent us some films from Guatemala taken at 120 PKV. I cannot answer your question in regard to that. We have to do some of the same people at this extremely high PKV and our conventional PKV.

In our senior citizens study, many aging hands look something like vacuole syndrome in this reduction in carpal space. You will find in most of the group in the 70's, 80's, and 90's a variety of clinical symptoms.

Dr. ROCKOFF. With regard to the carpal bone changes, one patient you showed had typical rheumatoid arthritis.

Dr. GARN. These carpal bone changes, however, appear in subjects which do not evidence signs of arthritis as well. In addition, the changes in the hand, in terms of cortical thickness, are parallel to changes in the first metatarsal, which bone, lacking a decent cortex, has to be explored purely densitometrically.

Now, there are three points I would like to make here. First, of course, is the over all reduction in cortical thickness in subjects of both sexes. We had hoped incidentally that the statement that people with rheumatoid arthritis did not lose as much bone would be true. It did not appear to be so.

Second, this change in both sexes appeared in all of the racial groups that we have studied.

Third is the question of amount of change. If we can extrapolate from the data we have here, the data we have on the foot, if parallel order of losses in Dr. Arnold's material may be considered, then one may project an estimated bone loss that, while considerably greater percentagewise in the female than in the male, 50% greater percentagewise, is of a comparable order of magnitude in both sexes. Rather than thinking of the male as not losing bone, it would appear from our studies that the amount of bone loss is the same. The percentage bone loss, however, is greater in the female who has the smaller skeleton to start with. In terms of the clinical implications of these findings of ours, I think we have to distinguish between those individuals who have little bone to start with and lose an average amount of bone and those individuals who were average or had above average amounts of bone to start with and then lose a considerable above average amount of compact bone.

Dr. ROCKOFF. The title of your talk was Normal Osteoporotic Bone Loss. Was your patient within the series of normal?

Dr. GARN. The word normal I am here defining in terms of people unselected on the basis of illness, history, or any other factor. In the case of the participants in our regular Fels studies, they joined the study at the time their first or their second child was 3 months *in utero*. They were unselected people, normal in that sense. Then, we have the word "osteoporotic" in quotation marks for obvious reasons because, although it is bone loss, there is the question of whether it is osteoporosis or not. Thirdly, we have the word "bone" in quotation marks because there is the question of whether your bone, my bone, and his bone, in terms of different types of bone, is the same. I think, Dr. Rockoff, you and I are probably in agreement that this is loss.

Dr. ROCKOFF. Yes, except you have an unselected series rather than normal. You can have a patient walk in with an amputated hand and that certainly is a bone loss. I think there has to be some separation of people who have obvious disease. The patient, if the uric acids are normal, has a diagnosis of advanced rheumatoid arthritis. To say this is a part of a spectrum of normal bone loss, I think, is wrong.

Dr. GARN. We have the fact that in her two sisters we had a comparable amount of bone loss. We have not yet had her brother in. I think we have the problem of whom we exclude as well as whom we include, which is the problem that I faced years ago in coronary studies where many people argue that one could not include anybody with a cholesterol over 280 as a "normal" individual. I think that as long as we define our "normal" growth in this sense, that while it is then possible to argue, as you have, about specific individuals, we do at least know what we are talking about in terms of material.

Dr. ROCKOFF. I don't think you have to stretch criteria very far to exclude from a normal study patients who have an obvious disease that can affect bone mineral.

Dr. NORDIN. I agree. It will, of course emerge in the statistical evaluation, unless half the patients in Dayton, Ohio, suffer from rheumatoid arthritis. These cases will provide a basis which presumably Dr. Garn will take into account when he establishes his normal values against age. I cannot believe that one value, three standard deviations off to one side, is going to be allowed by you to determine your normal means.

Dr. ROCKOFF. Dr. Garn described the changes in her wrist as occurring in many patients.

Dr. NORDIN. It is unfortunate he chose as his example a case of severe rheumatoid arthitis. I presume the others are normal?

Dr. GARN. I noticed this lady because of all those we had seen, she had the thinnest cortex. We are waiting for her to break her arm, also her two sisters probably will break theirs. I would like to point out that if you look at the bones of the older subjects, I do not feel that you are going to find more than 10% whom you would then call normal by usual clinical standards. The majority of the individuals over 65 have not only marked loss of bone material in the first metatarsal but tremendous enlargement of the sesamoids of the first metatarsal. In several cases the tarsal sesamoid grew to the walnut size in the X-ray. I had not known of the situation before we started looking at the feet of the aged. As soon as we start reviewing these older subjects, a variety of conditions appear in the X-rays. The question, then, is: Who should we throw out from such a group and are the ones remaining the normal ones for the point of our study?

Dr. SMITH. Dr. Garn, you mentioned a 23%, something like that, reduction in bone loss. Now are we talking about a 23% reduction in bone thickness because it is endosteal loss? That does not mean 23% reduction in the total amount of bone that is in that total hand, does it?

Dr. GARN. The 23% loss of thickness in cortex was not markedly different from the percentage loss if one expressed it as a percent of cortical area.

Comments on Cortical Thickness Measurements

RICHMOND W. SMITH, JR.
Division of Endocrinology
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I will define the population in answer to Dr. Rockoff's questions about rheumatoid arthritis. We did exclude the rheumatoids, the cortisone-treated patients, but they are a general outpatient population in this study of two thousand women.

I have here some data on the metacarpal cortical thickness mentioned by Dr. Garn 2 days ago, measured essentially in the same way, not at the mid-shaft but at the point of maximum cortical thickness.

In data plotted against age, Dr. Garn's Southern Ohio white females bear out his comment of 2 days ago that when independent workers use different methods, on the one hand, or calipers, we come to essentially comparable values for the mid-shaft of the second metacarpal. In regard to Dr. Nordin's comments as to any particular dip at a certain age, as to whether this is significant I do not know; I doubt it. It looks fairly linear with time.

Here we have plotted out the changes, the regression in the mid-shaft second metacarpal thickness for all subjects as against the regression for the same position on the Negro population, which are included in this group. That is, I have not taken the Negroes out of the total subject group. If we were to do this, it might shift this curve a little bit more and perhaps make the P value a little more impressive. We do see then a change in the Negro with age, but at a somewhat slower rate.

I have divided the regression of the mid-shaft metacarpal thickness into two groups based on visual estimated relative vertebral density. For whatever it is worth, when women with the good bone (that is, with grades zero and one, essentially normal vertebral density, which is comparable to the density seen in 25-year-olds by a series of studies we have made) are compared with the rate of loss in the metacarpal for the women with the bad spines (that is, those who show a loss of vertebral density 1/M again we are talking about the second and third lumbar vertebra, visually estimated), we can't say that there is a difference in the rates of loss in metacarpal thickness between the good spine group and the bad spine group.

In respect to Dr. Nordin's comments as to whether we have physiological osteoporosis and perhaps a pathological osteoporosis, we have the distribution of metacarpal cortical thickness for our symptomatic multiple fracture group scattered along the mean value for the survey group, that is, the asymptomatic ambulatory group. If we take two standard deviations which would be about 1 mm, we have women at age 60 who have very good metacarpals and we have osteoporotic patients who go well below the two standard deviations from the average value for the asymptomatic or so called normal population. Much more data obviously are needed.

In respect to the femur, we found in the two thousand subjects that with age there was a very definite regression of mid-shaft femoral cortical thickness in all subjects, black and white; we have the values for 132 Negro women. I think the irregularities are due to sample size, and not perhaps to any age significant variations. There is a difference between the two racial groups.

If one measures the leg in mid-position and

measures at the same point as one measures cortical thickness, one will find with age a very definite increase in the external or periosteal diameter. This appears to be greater for the Negro population. This perhaps is sample size; we have very few in this age group for the Negro women. But it is occurring in both and appears to be occurring more rapidly in the Negro female.

We have compared the expansion of the external diameter at mid-shaft, in 31 to 32 randomly selected subjects from this population group you have just heard about. In the subtrochanteric area the expansion was exactly one-half of the mid-shaft, and the femoral neck, again read in the same film, showed less than a millimeter increase. Our suggestion is that perhaps with age or with continuing weight bearing there is a flexion in the mid-shaft which activates subperiosteal bone accretion. The significance of this, of course, to osteoporosis is not known at this time, but one can see that if this continues substantially, it makes the neck vulnerable to fracture if it is becoming more rigid due to mid-shaft expansion.

COMMENTS

Dr. TROTTER. I would like to ask Dr. Smith how much range there is in the site of greatest diameter or greatest width of the cortical bone in the shaft. Is there much range or is it in approximately the same place or level of the shaft?

Dr. SMITH. When you measure the whole femur, maximal cortical thickness is essentially at mid-shaft, but actually our X-rays cut off; we did not look at the lower femur.

Dr. URIST. I think we can confirm that observation. We have done this with microradiographs. There is a layer of bone in the severe osteoporotics that is laid down and it is not Haversian, it is lamellar. Therefore there is a qualitative difference in the bone that is laid down.

Dr. SMITH. You are speaking of circumferential lamellar bone.

Dr. URIST. Yes. We have shown this in our radiographs. I don't think any of us appreciated that it was measurable until you demonstrated it.

In your slide on aortic calcification, this is just indiscriminate calcification, this is in the elastica lamella. This is the specific thing that increases with time. I do not know whether it is related to physiologic osteoporosis or pathologic osteoporosis, but it is a definite process that increases with time. Bayer, Hastings, and Lowry measured this chemically in unselected cases at autopsy.

Dr. SMITH. I think it might suggest that there are changes going on in connective tissue of the structure of the bone.

We made a rough gradation of the degree of aortic calcification, not whether it is present or not. We used an arbitrary scale, one to four. What I have presented is accumulated scores for the grade zero and one, that is, good spine patients matched against the more osteoporotic, and again asymptomatic osteoporotic women versus the more severely osteoporotic. We have done this now for all two thousand patients. Then, for the three 10-year age groups we have given the relative scores of aortic calcification on the basis of one to four. I can't do it any better than that. I can't weigh it, I don't know how to measure it, but as the subjects get older, the differences narrow. These are the relative values, and I can't carry the interpretation further than that.

Dr. URIST. I think Dr. Babcock's presentation earlier was a good demonstration that Dr. Cameron's technique can be used clinically. This is an apt subject for presentation in this symposium for NASA since here is an experimental animal that demonstrates pathological osteoporosis because these are young animals. They also demonstrate the effect of immobilization and confinement in a cage which would be comparable to the couch where the astronaut would have to sit in for a month at a time. I first became acquainted with this condition in chickens when they brought me some of the specimens, and I found that they had low bone mass.

This was very exciting because the bone mass increased with exercise. This is not a problem which has anything to do with aging. I changed the name to cage layer osteoporosis because the bones were thinner, they were more brittle, they were light. The cortical bone was eroded from the inside only, and the bones did not respond to any known treatment.

Dr. ROCKOFF. I would like to make some remarks about general experimental design and applicability of the various methods that have been mentioned so that we can all talk in the same terms. First of all, I think that most of the misunderstandings that have arisen have been in the interpretation of data, because people used different means for selecting their populations and they were then working with markedly dissimilar systems. People have been talking in terms of accuracy and precision as applied to half centimeters in the case of hens, and many, many centimeters in the case of human vertebral bodies. I think these differences have to be stressed when people talk about one technique and whether it looks as though it might be potentially better than another. It has to be defined in terms of what parts of the body are being dealt with.

These are just random unrelated comments, but Dr. Mack, with regard to your system of replication, you

said you do not do more than one film because of the radiation dosage.

I would suggest that the dosage level with a couple of millimeters of added aluminum filtration for the calcaneous with a fairly well collimated beam is about 0.03 r. If you do a film a day for 30 days you are still less than 1 r. Considering that bursitis of the shoulder used to be treated with 75 r a day for 3 days, we are talking about dosages that have been shown to do nothing deleterious to the human body.

Dr. MACK. Where we were doing the os calcis and the finger, we did more films, but when we added other bones, we cut down to one and replicated the measurement of that one film. I am sure you are correct. Earlier we did do more than one film on the two small bones.

CONFERENCE CONCLUSION

Concluding Remarks

G. Donald Whedon
Dale W. Jenkins

Dr. Jenkins. I should like to congratulate the speakers and the chairman this morning for keeping to the schedule, even though we had a very full and complete schedule. I now shall call on Dr. Whedon for concluding remarks.

Dr. Whedon. Thank you, Dr. Jenkins. I think that perhaps the most significant thing I could say at this time is that from the experience we have had over the past 2½ days no very great or weighty comments need to be made.

We have certainly had an enjoyable and exciting time. When Dr. Jenkins and Dr. Neuman and I conferred by telephone and letter to select this program and the participants, as I indicated at the beginning of the conference, we purposely tried to obtain people from as many different disciplines as possible in order to develop interaction between individuals of different training and different scientific backgrounds. Obviously, the problem of bone densitometry requires the combined and collaborative efforts of people with many different sorts of training and experience.

I noted through the conference a predicted language or communication difficulty among people with different backgrounds. But I also noted some evident problems, I would even say mistakes, in planning studies and in procedures in studies when individuals with one background of training and education undertook work in a quite different area of discipline. There was here and there through the conference an indication of inappropriate techniques used and inappropriateness of expression of the data that they obtained. I noticed this, of course, particularly in the individuals originally trained, let us say, in the hard sciences or the nonmedical disciplines in taking up biomedical studies. But that is undoubtedly because I am on the biomedical side, and I suspect that others noted the same business in the reverse direction when those of us having medical training tried to get into more mathematically oriented or physically oriented projects.

I think a lesson we might draw, a suggestion I would make, very cautiously and as tactfully as I can, is that those of us who are going to continue to do studies of this nature in which we shift over into another discipline, should make very serious efforts to obtain sound advice from other individuals in the new fields that we are going into before the project ever begins. In this way we will get the most for our effort; when we come to the end, we will have a better chance of obtaining a meaningful result, and we will be able to present our data in a way which will be most easily communicated to other individuals. I say this with a very optimistic feeling that the conference has been so successful that we shall have to have another one. How soon and whether it will include precisely the same individuals and be on precisely the same techniques, I am not sure. It will take some time to decide. But obviously bone densitometry in the broadest sense is very active and is propagating very rapidly. It will be soon time, I think, to come back and meet again, perhaps not in this room or under these auspices but under some auspices. Thank you all very much. I am so glad that you enjoyed yourselves.

Dr. Jenkins. This has been one of the most

vital, interesting, and sometimes exciting working conferences that I have attended. The conference has been of extreme value to NASA in looking into present techniques and those which are being developed, and in helping us assess their potential. It will help us in deciding what courses we shall follow in our future studies in NASA in investigating the skeletal and muscular problems in relation to weightlessness and in defining just how serious a problem this is.

It is gratifying to find general agreement and resolution of most of the major controversial problems in bone X-ray densitometry. It is a real accomplishment to have the accuracy and precision of this method clearly defined and the magnitude of possible error accurately quantified. This is a worthwhile advance and will be of great value in experimental studies in the Biosatellite, Gemini, and Apollo Programs. This conference has also pointed out the difficulties and need for more research in the area of correlation of change of X-ray film density with change of bone mineral. Another problem that has been identified and requires more research is the correlation of change in mineral content of measured bones with the total change in the skeleton.

On behalf of NASA, I thank all of you for interrupting your busy schedules to participate in this conference. We appreciate your submitting manuscripts as requested, and the editors will edit the proceedings as fast as possible. We appreciate the remarks of most of the participants that this conference has been of great value to you. May I conclude by saying that the conference has surpassed the expectations of the organizers, and will be of lasting value and importance to the sponsors.

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